

NJSE

Nigerian Journal of Science and Environment

*Official Publication of the Faculies of Science and
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Editorial Comment

The tenth volume (Nos. 1 & 2) of the Nigeria Journal of Science and Environment (NJSE) which is a special issue of peer reviewed papers presented at the the Delta State University Faculty of Science Maiden Conference (FOS2010), marks another important milestone in the publication of the Journal. Since the birth of the first volume in 1988, the Journal has continued to improve both in quality and speed of publication of submitted articles. This is because of the commitment of our reviewers and members of the

editorial board. The editorial board is pleased that this issue is in print within the year of expected publication, 2011.

Our esteemed contributors will be pleased to know that NJSE has been on-line since 2007. This means that manuscripts for the Journal could be sent electronically either by disk in word format or by e-mail to the Editor. In addition, all subscribers will be able to access the Journal on-line and receive instructions automatically when their subscription is processed. We assure all our contributors that there will be regular publication of the journal henceforth.

The Nigerian Journal of Science and Environment will continue to make the results of the research of members of the Faculties of Science and Agriculture as well as other reputable scholars outside our University available to intellectual community. To this end, this issue embraces publications from participants at the FOS2010 within and outside Delta State University.

The Editorial board want to congratulate the authorities of Delta State University for their financial support for the Journal and the Faculty of Science for the successful conference and its financial support for this issue.

The Editorial Board of the Nigerian Journal of Science and Environment joins the Guest Editors in appreciating our referees for their effort in reviewing the papers in this special issue. A particular debt is also owed to the Associate Editors for accepting to serve.

Enquiries and articles should be sent to:

Prof. Patrick N. Okoh

Managing Editor

Nigerian Journal of Science and Environment c/o

Department of Biochemistry,

Delta State University, Abraka, Nigeria.

Or to

Assistant Managing Editor

E-mail: asagbabch@yahoo.com

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Nigerian Journal of Science and Environment

AIMS AND SCOPE

The Nigerian Journal of Science and Environment publishes original pre-reviewed research manuscripts in agriculture and pure and applied sciences. The Journal is designed to contribute towards the promotion of science particularly in the developing countries of the world. Manuscripts on environmental science with particular reference to the African continent are highly welcome.

The Subjects covered include:

Agriculture
Biochemistry
Biology
Botany
Chemistry
Environmental Science Geology
Mathematics / Computer Science
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Physics
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TYPES OF CONTRIBUTIONS

Original research papers; review article; technical notes; book review; reports of conferences and meetings; short communications; letters to the Editor.

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Manuscripts should be in doublespaced typing in A4 paper with a wide margin to the left handside. The title page should contain the title, author(s) name(s) and affiliation(s), including a complete address for

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The SI units should be used for all scientific and laboratory data; if in certain instances, it is necessary to quote other units, these should be added in parentheses, temperatures should be given in degree Celsius.

The abbreviations for units should follow the suggestions of the British Standard publication BS 1991. There is no need to use full stop to mark abbreviations, e.g. M (not M.) ppm (not p.p.m)

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BOOKS

Smith, J.D. (1984); *Introduction to Animal Parasitology*, 3rd Edition, Cambridge University Press, Cambridge.

CHAPTERS IN EDITED BOOKS

Jeffries, P. (1994). Biochemical cycling and arbuscular Mycorrhizas In: the sustainability of plant-soil systems, *Impact of Arbuscular Mycorrhizas on sustainable agriculture and natural system*. Gianinazzi, S. and Schuepp, H. (Eds.), Birhauser Verlag, Basel. Pp 101 – 115.

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HANDLING CHARGES

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**Welcome Address by Professor S.H.O. Egboh, Dean, Faculty of Science,
Delta State University, Abraka, at the First Faculty of Science Conference (FOS
2011) At Pre-Degree Hall on Tuesday, 2nd February, 2010**

It is with grate pleasure that I welcome all participants on behalf of staff and students of the Faculty of Science to the Delta State University, Abraka for the First Faculty of Science Conference tagged “FOS2010”. Today marks another great day in the annals .of the Delta State University, since this is the first time that the Faculty is hosting its own conference since its establishment in 1992. Today will go down memory lane as one of the most important days in the history of the Faculty, for it is the first time we took up the challenge to host an event and showcase our research endeavours. The faculty wishes to assure all that the Annual Conference will be sustained. In addition, the proceedings of the conference will be published regularly.

The theme of this year’s conference is *INVESTING IN SCIENCE: GROWTH AND SUSTAINABLE DEVELOPMENT OF DELTA STATE*, and the following are the subthemes: *Biotechnology and Biodiversity, Earth and the Environment, Toxicology and Human Health, Physical Science and National Development, and Science and Mathematics Education*. The importance of investing in science to national development cannot be overemphasized. The choice of the theme is apt due to its importance to national development, especially in these days of global economic meltdown. The major characteristics of this depressed economy include downward movement of prices of goods and services due to falling aggregate demand within the economy and declining level of general output due to lower productivity and low elasticity of supply. Considering the complexity of the forces operating in global environment and the corporate strategy adopted by nations and organizations to stimulate growth and develop the capacity for science and innovatiron, the hosting of such conference is timely.

Nations and organizations are expected to utilized the opportunitbity presented by the current economic meltdown to improve the nations by exploring areas of competitive advantage that could be leveraged upon to sustain the emerging economy. No national economy can grow by neglecting innovative science and technology. Thus, we need to encourage developmental ideas that can change the fortunes of the economy. Sustainable development requires the exploitation of resources, direction of investment, orientation of technological development and institutional changes which are in harmony and enhance both current and future needs and aspirations. This involves meeting the present needs of humans without endangering the welfare of future generations. Thus, it is known that economic, social, political and environmental sustainability factors determine the overall sustainable development.

Development should be complementary to environment and should ensure search for better investment climate or market, challenges to better education, improvement of quality of life, etc. The current global economic meltdown has been a source of serious challenges to scientists, individuals, businesses and countries. This period has brought sudden economic hardships to people, organizations and nations. Organizations have had a scale down operations and cut down on their workforce. What roles should scientists play in providing solutions to these challenges? What preparations and plans should we make towards a better future for our people? This conference would attempt to provide answers to these and other questions.

The level of unemployment and underdevelopment among scientists is embarrassing considering Nigeria's divine endowments, especially in oil, gas, minerals and other resources. The acquisition of appropriate talents/skills by citizens is critical for our nation's sustainable development and global competitiveness. The theme and subthemes are apt as efforts are being made to refocus, reorientate and reposition science to prepare scientists and allied disciplines for the challenges of self-reliance and thereby create employment, reduce poverty and generate wealth. The conference would be of great benefit to you, and you are therefore expected to participate actively in all the activities of the conference. The conference will also afford the participants the opportunity to brainstorm and proffer to the ills plaguing the teaching, research and development of scientific principles, as well as help develop workable reforms and innovations.

We are grateful to our invited guest speakers for accepting our invitation to participate at the conference. The faculty acknowledges the financial and materials assistance of the Delta State Government, Delta State University, Rector of State Polytechnics, individuals and several organizations towards the hosting of this conference.

I wish to assure you that your stay in Abraka will be a memorable one. Finally, I wish all participants successful and peaceful deliberations and fun while the conference lasts. You are, indeed, welcome to Delta State University, Abraka.

Thank you, and remain blessed.

Professor S.H.O. Egbah
Dean of Faculty of Science

Guest Editors' Comment on this Special Issue

The Faculty of Science of the Delta State University, Abraka (Nigeria) held its first annual conference from February 1 - 4, 2010 with the theme: *Investing in science; growth and sustainable development of Delta State*. There is general consensus today that the classification of countries into developed and developing nations depends on their level of acquisition of science and technology (S & T). This is because irrespective of the country's history, resources or geographical location, it is her ability to harness her resources (human, natural and artificial) that determines its economic success. The power to harness these resources depends largely on the level of acquisition of S & T.

It is widely believed that Nigeria is blessed with both human and natural resources that will make most economic models very successful. Our present level of development, however, is partly due to our inability to harness these resources. Interestingly, there are reports now and then of our country men and women that are excelling in various fields of S & T in various parts of the world and thereby contributing to the economic growth of these countries. There are also reports of promising scientific scholarship of scientists in various parts of our country who, however, are struggling against poor scientific funding to forge ahead. Therefore, appropriate investment on the scientific community will lead to integral growth and sustainable development. The objective of the conference is to mobilize the scientific community to package this clarion call. Thus the sub-themes were broad to cover various aspects of science such as Biotechnology and Biodiversity, Earth and the Environment, Toxicology and Human Health, Physical Science and National Development, and Science and Mathematics Education.

There were two invited speakers and a total of 87 contributed presentations were made under parallel technical sessions for the physical sciences and life sciences. Many of the participants sent their full papers after the conference and after a peer review process, the ones published in this special issue were accepted. This special volume is a double issue comprising Volume 10, Nos. 1 & 2. The reason for this is that the papers from both the physical sciences and the life sciences are more than the editorial content of a regular volume of this journal.

We wish to express our deepest thanks and appreciation to the Editorial Board of the NJSE especially the Managing Director, Prof. P.N. Okoh and the Assistant Managing Editor, Prof. S. O. Asagba, for their commitment and assistance in seeing this issue to its fruition. We also deeply appreciate all the referees for their insightful and constructive comments which have made the papers in this issue more valuable.

On behalf of the Organizing Committee of the conference (Prof S.H.O. Egboh – Chairman, Dr. N.J. Tonukari – Vice Chairman, Dr. G.E. Akpojotor – Secretary, Dr. C.E. Mokobia, Dr. E. Adaikpo, Dr. O. Kori-Siakpere, Dr. U.B. Owhe-Ureghe, Prof. S. O. Asagba, Dr (Mrs) N.E. Edema, Dr. J. N. Igabari, Dr. E. C. Okolie and Dr. C. M. A. Iwegbue, we wish to use this medium to acknowledge the Vice-chancellor, Prof. Eric Arubaye; the Commissioner for Education represented by his Special Assistant; the Commissioner for Science and Technology represented by the Permanent Secretary, Dr C. O. Ukpe, Deputy Vice Chancellor (Administration), Prof. (Mrs.) O.C. Okobia; Deputy Vice Chancellor (Academic), Prof. R. B. Ikomi and other members of the university management as well as our invited speakers, Prof. Philip Kuale and Prof. Ogi Okwumabua, for their various roles and support in making the conference a huge success.

It is also pertinent we specially appreciate all members of the Organizing Committee for their various contributions to the success of the the conference. Thanks and appreciation are also due to all the faculty members and to all the participants.

Finally, to make it special, we decided to conclude by wholeheartedly acknowledging and appreciating the commitment and tireless effort of the Dean of the Faculty, Prof S.H.O. Egboh, to make this conference successful and that this proceedings is published.

Godfrey E. Akpojotor
Nyerhovwo J. Tonukari

INVESTING IN SCIENCE: GROWTH AND SUSTAINABLE DEVELOPMENT OF DELTA STATE

Ogi E. Okwumabua

Dept. of Pathobiological Sciences/WVDL, 445 Easterday Lane, Madison, WI 53706; USA
Phone: 608-262-4168; e-mail: ogi.okwumabua@wvdl.wisc.edu

Abstract

The classification of countries into developed and developing nations are results of levels of acquisition of science and technology. Studies and evidence have shown that the differences in education, scientific and technological infrastructure and in the popularization of science and technology in the two groups of countries are the most important causes of differential social and economic levels. For example, the key factors that contributed to the economic success of Japan, USA, Canada, Germany, United Kingdom, and China amongst others are recognition of necessity for good education and sustained and aggressive investment in basic scientific research and technology, manpower and infrastructure. In effect, the countries have achieved remarkable innovations in Agriculture, electronics, health sciences, biotechnology, engineering and information technology which in return have dramatically raised the quality of lives, increase productivity of business, and created long term economic growth. A major obstacle to development in the developing nations and states remains the lack of scientific and technological capacity due to lack of investment in the broad area of science. In the last decade poor states in the developed countries such as the USA began to realize that they must place heavy reliance on education in seeking solutions to their formidable economic problems. Because science education is the primary engine for economic growth and the key to unlocking any state or country's potential, Delta State should make honest effort to invest in science particularly in the areas of infrastructure development, provision of quality education, technology adaptation and improvement, innovation and development and policy implementation. Thus the state government must commit to having sizeable annual budget sufficient for capacity building and enhancement. Science-based growth and sustained development of the state can be achieved if the state government support transformation of science education and provision of excellent opportunities and healthy environment for teachers and students to engage in the most, cutting edge research.

Introduction

The social and economic growth of developed countries is dependent on an essential emphasis on education, science, and technology. The basic problems of developing countries are the weak education and scientific infrastructure, and a lack of appreciation of the importance of science as an essential ingredient of economical and social development. Inadequate scientific infrastructure is a critical factor which creates strong barriers to the path of advancement in developing countries.

Today, in developed countries, basic and applied scientific research is an essential investment and nurturing scientific and technical talent, and to the concomitant training of students. What is emerging from this priority is the close association of education and economical growth. Accelerating the rate of growth and rate of productivity can basically be accomplished by stimulating and supporting scientific education in universities. Salam (1989) states that science in developing countries has been treated as a "marginal activity" and perceived even as "ornament". Indeed, most of the developing countries do not realize that their situation can only be rectified with the infusion of modern science and technology into their societies.

The differences in the scientific and technological infrastructure and in the popularization of science and technology in the two groups of countries are the most important causes of differential social and economical levels. An essential prerequisite to a country's technological progress is early recognition of necessity of a good educational system. This was one of the key factors that contributed to Japan's economic success. The role of Technion, the Hebrew University of Jerusalem and the Weizmann Institute in Israel's rapid development cannot be underestimated.

Modern science permeates every aspect of economic and social life. For this reason, education, research and technology as instruments for accelerating development should receive special attention in national planning in the developing countries. In order to make realistic plan, not only a vision, but also scientific leadership, and investment in scientific enterprise both by government and private sectors are required.

Turkish industries expect engineering graduates to have the current know-how to solve immediate problems. This expectation is often reflected in university curricula: there is a tendency to teach as many courses as possible in the core subject. A sustained and aggressive investment in basic scientific research, manpower and infrastructure is needed, like that triggered by Sputnik or devoted to the Manhattan project.

Basic science and technology have given us remarkable innovations that have dramatically raised the quality of our personal lives, increased the productivity of our businesses, and created long term economic growth. The challenges we face today will require new generation of inspirational breakthroughs from basic science to replace the economic recession with economic growth, to replace uncertain and costly imported oil with a secure and sustainable energy supply, and to reduce carbon dioxide emissions that threaten global climate.

Global Perspectives

We know that many of the next generation sustainable energy technologies are: carbon capture sequestration, high-efficiency coal and nuclear electricity; renewable solar, wind and geothermal power generation; solar fuels and biofuels; solid state lighting; energy storage for plug-in hybrid and battery electric cars, and high-temperature superconductivity for 21st century electric grid.

1980s: MIT, Stanford University and Silicon Valley, North Carolina and research triangle began to receive considerable attention.

Therefore in the 1980s, there was emergence of state science and technology programs with direct links to economic development in the United States. States such as North Carolina, Connecticut, Georgia, Pennsylvania, Ohio, and New Jersey pioneered an increased state economic development focus on technology.

States such as North Carolina, Connecticut, Georgia, Pennsylvania, Ohio, and New Jersey pioneered an increased state economic development focus on technology.

A Minnesota study found that states spent more than \$550 million on state science and technology efforts in FY 1988 alone. This same study found that 68% of state funds were used for research (27 states) or technology development centers (29 states), 7% for business start-up support (20 states), 8% for technical assistance problem solving (23 states), and 17% for technology education and training (4 states).

The Michigan Technology Deployment Service: Established in 1985 to assist companies that are considering adoption of new computer-based manufacturing tools and methods.

The Ohio Technology Transfer Organization: provides Ohio business with direct access to new technology and research through a statewide network of thirty-four technology transfer agents based at two year colleges.

Maryland operates regional technology extension offices in conjunction with the Engineering Research Center at the University of Maryland.

The Washington Technology Center

Pennsylvania's Ben Franklin Partnership Program

To meet the challenge of economic competitiveness, the United States as a nation has been making a long-term commitment to its education and research system. Science and engineering research and education have in the past, and will continue in the future to play a crucial role in determining U.S. competitiveness (DeLauro, 2008).

A healthy population is an absolute prerequisite for sustainable development; the individual, societal and economic consequences of infectious diseases are seriously inhibiting development in many countries.

Vaccine research in the prevention and control of both communicable and non-communicable diseases has made a vital contribution to national and International health development during the 20th century. Technology in areas of public health such as disease control, medicine and medical electronics needs to be developed. Another area is the protection of the environment for better dwelling conditions on the one hand, and for increased productivity of the land on the other.

Medicine is already being transformed with new genetically engineered drugs and non-invasive surgical procedures. For example, today we can slip new genes into plants and animals and create new drugs. In agriculture, we can now create pest-resistant crops and engineer desired flavors into fruits and vegetables. In physical sciences, continued advances in molecular engineering are expected to result in highly specialized materials for all sorts of uses. Nanotechnology promises miniature robots that could crawl around in our arteries, remove clogs and perform surgery. The purpose of all these is to have improve quality of life.

What can we expect from biotechnology? In the last 20 years, biotechnology based upon recombinant DNA has developed invaluable new scientific methodologies and products in food and agriculture. It is a new technique, not a new system that has been added to plant and animal breeding. Gene alterations have conferred producer-oriented benefits, such as resistance to pests, diseases, and herbicides. Other benefits likely to come through biotechnology and plant breeding are varieties with greater tolerance of drought, waterlogging, heat and cold-important traits given current predictions of climate change. In addition, many consumer-oriented benefits, such as improved nutritional and other health-related characteristics, are likely to be realized over the next 10 to 20 years. Insect resistant cotton is grown in China, India and South Africa, where it significantly reduces production costs and increased profits. Growing potential of science to improve the nutritional quality of our food supply.

Science and technology has been used and will continued to be used to fight terrorism and crimes in general. Needless to state, sustainable development in the absence of security is almost impossible. This why many developed countries have also dedicate some funding to to initiate research and development activities in these critical areas. So indirectly it is observed again that Technology is the primary engine of economic growth and provides the key to unlocking any country's potential. Hence countries that want to develop must invest significantly in science and technology.

Transition to information society is a necessity for global competitiveness. This transition to information society will demand the development of information-related industries centered around micro-electronics, communications, computers, etc. Moreover, reducing the labor component of production systems through automation technology will require re-educations of displaced labor.

The long-term goal of science and technology should be in accordance with that of national development. It should include adopting a strategy for industrialization and recognizing the central role of education in efforts to stimulate prosperity. Further, measures have to be put in place to motivate scientists so that they

are retained in their countries to reduce brain drain. This can be achieved by continuous and increased government support for S&T to meaningfully contribute to socio-economic development.

Institute capacity-building program in emerging technologies such as biotechnology and information technology and put in place policies geared to promoting their development and application. Teaching of science should be done at all levels of education. Technical education must be improved to cope with technological changes. R&D on high priority areas should be emphasized. S&T programs should aim at wealth creation. Business incubation centers should be established. I read in Animalu and Akpojotor (2009) that Nigeria accompanied this feat in the 1990s when the Technology Business Incubator (TBI) centers were established at Agege (Lagos State), Aba (Abia State) and Kano (Kano State) by the National Technology Foundation (NTBIF). Therefore, the Delta State government as well as researchers at the DELSU can liaise with these centers, if they are still viable.

Countries should endeavor to establish stable and effective S&T institutions that should provide clear policy direction. Universities have been the principal stimulus to electronic and science-based industries in California, North Carolina, and the New England States. Poor state in the US began to realize that they must place heavy reliance on education in seeking solutions to their formidable economic problems.

It follows then that to meet the challenges of the 21st century, Delta state must rely heavily on education at all levels for creation of the type of cultural environment which now weighs so heavily when government and industry select sites for laboratories and manufacturing plants. First-rate public schools and year-around recreational programs for youth are given high priority in community evaluations. I will elaborate more on this later when considering how to train the next generation of scientists and other professionals in Delta State.

Still on the global perspectives, it is important to re-emphasized that science and technology reduces poverty and achieves sustainable growth (says World Bank president Paul Wolfowitz). If you want to tackle poverty, science technology and innovation must be part of the picture. Third world countries must build their scientific and technological infrastructure, by seeking loans from the World Bank.

Expanded scientific activity is thought to benefit national economic development through improved labor force capacities and the creation of new knowledge and technology. Effect of scientific infrastructure and national economic growth are examined.

Innovation is driver for economic growth. Innovation based economic development (IBED) supports the alignment of the various inputs required for innovation (research infrastructure, technology transfer, start-up capital and businesses support, etc. Example drawn with Korea, Singapore, and Taiwan (these countries are challenging traditional innovators like Japan, Scandinavian countries and the United States).

In today's knowledge economy, innovation has the power to galvanize industry and drive economic growth, thus creating the knowledge intensive, high application, jobs that are the ultimate objective for today's economic development community.

India: growth has been driven by India's pipeline of engineers and computer scientists, primarily a function of its strong training and education programs in these areas. The Indian Institute of technology is known throughout the world for producing high quality engineering and computer science graduates.

Finland; has established itself as global leader in innovation, particularly in the area of telecommunications including the world's first Global System for mobile communications (GSM) network and has produced world-class companies in the industry such as Nokia.

The success story of the emergence of Israel from a 'scattered nation' to regroup in the desert to become a powerful country epitomizes success in innovation driven by high levels of R&D.

A proactive and relevant education plan built on an aggressive infrastructure development. Therefore an educated workforce is critical to the success of the new economic development plan. Talking about infrastructure development plan, it must be emphasized that to stimulate economic development, governments must either invest in, or stimulate the private sector to invest in the development of well-equipped universities, community colleges, trade schools, high schools and elementary schools. Further, there should be an excellent intellectual property System. This will promote competency to discover, assimilate, utilize, learn, and improve new industrial technology, which is the key determinant of successful development of local manufacturing sectors. There should also be a normal innovation system as this will promote effective technology development in a country depends on the network links among private and public institutions that shape technological capabilities.

Solutions to the major sustainability problems of the 21st century, including poverty alleviation, food security, health, a looming water crisis, decoupling of economic growth and environmental impact, renewable energy sources, desertification, diminishing ecosystem services, biodiversity maintenance and use, climate change, and the rise of megacities-all critically require knowledge from scientific research and appropriate technologies (Animalu et. al, 2005).

The message from the global perspectives is very lucid: a major obstacle to sustainable development for much of the world remains the lack of scientific and technological capacity, especially in developing countries. This has to do with poor funding for S & T. Therefore the roadmap to reverse this trend is to properly invest in S & T. The first step will be in capacity building which is a critical element for sustainable development. The remaining part of this paper will be on this capacity building.

Educating the next Generation of Delta State Scientists and Related Professionals

From the presentation so far, the need to invest on science and technology are obvious and an effort to make a cursory study of the path that has been taken by the United States and other developed countries as well as emerging economies have also been highlighted. As stated above, that the first step in investing in science and technology is to develop an adequate manpower and expertise. Therefore, I will summarize my invited talk presented at the inauguration of the Delta Diaspora Direct (D3) in New York City (USA) in September, 2009.

Scientists are individuals with expert knowledge in natural and physical sciences. These individuals learn, discover, invent and create important solutions that have an impact on regional, national and global level. Thus, the core of science is simply turning ideas into reality. Because science education deals with sharing science content and process with individuals not traditionally considered part of scientific community, educating next generation of Delta State scientists requires early exposure to the field and activities that impact knowledge and skill to those with interest in science and related disciplines. The framework to achieve these activities are governed by availability of acceptable infrastructure, adequacy of resources, qualified and dedicated personnel, government support and commitment, industry support, private and public support.

Infrastructure: Environment conducive to the learning of modern science and technology and infrastructure to conduct innovative research on critical scientific problems has to be in place and must meet the minimum acceptable standard required for the specific science discipline. These include the basic physical and organizational structures such as buildings (classrooms, laboratories, libraries etc), uninterrupted power supply and constant source of good water quality. Expert scientists must therefore be involved in planning and advising as they are most familiar with requirements and standard. The current status of infrastructure in Delta State is unacceptable as evidenced in Figure 1 when compared to my laboratory in my university in the US as depicted in Figure 2.

Resources: Science education provides understanding of the terms and techniques through “hands-on” experience supported by lectures integrated into the practical component. The combination of theoretical and practical aspect provides solid foundation into cutting-edge research developments and techniques which in turn leads to economic growth and development. Thus, there must be adequate resources such as a library rich in modern text books, manuals in practical and experimental science, journals and periodicals, audio-visual resources and computers with high speed internet capabilities. The key for training scientists lies on exposure of science students to techniques using state-of-the-art equipment. Therefore, laboratory facilities that are well furnished with modern instruments, equipment, reagents, materials and supplies are an absolute necessity.

Personnel: Having sufficient number of well trained personnel is of paramount importance. Most of the science educators in Delta State are “text book” teachers with little to no “bench top” training as such they are unable to teach the technical aspects of science. The lack of technical knowhow and research capability hinders economic development and growth. Thus, the State need qualified individuals who can mentor in the class room and laboratory to ensure that knowledge and use of modern methods are transferred from one generation to another; otherwise technological challenges of the future will remain unmet. This can be achieved by re-training current “science teachers” on the practical use and application of modern technology in solving scientific problems. This concept of “training the trainer” has significant merit because a scientist is only as good as the training received. “Training the trainer” can be achieved through talent tapping program in which visiting scientists with demonstrated evidence of accomplishment in theoretical and applied science are periodically invited to serve as mentors. Scientists can also be recruited on a temporary or permanent basis to come to Nigeria to provide “hands-on experience” mentoring to select individuals. Collaborative arrangements and exchange programs with renowned institutions within and outside Nigeria including the United States of America should be set up to facilitate science teacher and student training. Finally, Delta State scientists, students and citizens interested in the sciences should be encouraged and supported to attend seminars, conferences and workshops that deal with scientific issues.

Curriculum: Curriculum has to be descriptive, realistic, and directed toward meeting stated objectives. Additionally, it has to be focused and continually reviewed and updated.

Training and education: It has been shown over and over that interest and early education are keys to training future scientists. Therefore, starting from junior secondary school, students should be exposed to basic and applied science through lectures and simple hands-on laboratory experiments while participating in a variety of formal and informal science programs. This will stimulate their interest (basic law of learning) and prepare them for university education in the sciences.

At the University and other levels, the tendency for science educators to teach by charts alone without the use of actual apparatus and specimen should be discouraged. Rather emphasis should be placed on teaching basic and applied sciences that utilizes modern scientific equipment and methods for research and development which are crucial to the long-term health of our state. Because people improve their scientific knowledge through doing and participating in formal and informal interaction, industrial attachment in government agencies, private and public sectors and involvement in professional organizations should be encouraged.

Budget: Educating the next generation scientists will not come without money for infrastructure development including facilities, equipment, materials and recruitment and maintenance of personnel. It will also not come if money is unavailable to transform science teaching while providing excellent opportunities for teachers and students to engage in the most, cutting edge research. Thus, the state government should honestly commit to science education by providing annual budget sufficient for operational needs. Funding for basic research can deepen the knowledge base of society while educating the next generation of scientists. In developed countries, government-sponsored basic research is increasingly recognized as a key component of a knowledge-based economy. Other sources of funding

could be secured through public and private sectors but the government must pay its share in order to make others follow suit.

Summary:

- To educate the next generation of Delta State scientists and related professionals, there must be acceptable infrastructure to suit the need of each science discipline. These include buildings (classroom, laboratories, and libraries).
- There must be adequate resources such as modern text books, manuals in practical and experimental science, science journals and periodicals, audio visual resources and computers with high speed internet capabilities, laboratory equipment, materials and supplies.
- The current science teachers must be retrained while recruiting to increase the number of expert science mentors.
- Mentoring must start at the secondary school level to stimulate interest in science.
- Government must commit money for science education by having an annual budget sufficient for operational needs.

Conclusion

The US is a leading World Power today because of its investment on science and technology. This was recently asserted by the U.S. Rep. Rosa DeLauro (2008) that “Economic experts have concluded that science-driven technology has accounted for more than 50 percent of the growth of the U.S. economy during the last half-century. We have a responsibility to continue investing in that science so that it will pay more dividends in the century ahead. That is why the House Democrats’ Innovation Agenda calls for a doubling of the National Science Foundation and the Department of Energy’s Office of Science budgets over the next 10 years.” This has been one of the strategy to surmount the global economic meltdown and the emphasis has been on investing more on science and technology. Suffice it to conclude that for Delta State to meet the challenges of the human development index (HDI) whose three components are longevity (health), knowledge (education) and income (purchasing power), it must begin to invest in science and technology now especially as there is oil wealth in the state for this investment.

Acknowledgment

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Microbiology Laboratory, Delta State University, Abraka (January 2009)



Fig. 1 (a) (colour online): Microbiology Laboratory, Delta State University, Abraka (taken January, 2009)



Fig. 1 (b) (colour online): Microbiology Laboratory, Delta State University, Abraka (taken April, 2009)

An essential component of learning science is the process of hands-on-experience (active) rather than simply reading or watching (passive). Picture from the Diagnostic Microbiology at the University of Wisconsin, Madison (USA)



Picture from the Diagnostic Microbiology at the University of Wisconsin, Madison (USA)



Fig. 2 (colour online): Diagnostic Microbiology Laboratory, University of Wisconsin, Madison (USA) (taken during the visit of Dr Godfrey E. Akpojotor to the laboratory in March-April, 2009)

INVESTING IN SCIENCE AND TECHNOLOGY: it is overdue for our Socio-Economic Development

P.A. Kuale

Delta State University, Abraka,
Oleh Campus

1. Introduction

“We have left undone those things which we ought to have done and we have done those things which we ought not to have done”. This is how we have done many things to the detriment of the nation and have left the funding of science and technology adequately since independence, 1960 to date. It is true there has been some funding but to a very large degree, it has been grossly inadequate. Perhaps, because we do not know what it really means to fund research and development.

During this lecture, I will draw from some of the enlightened teachings which have been coming to mankind; and most of the teachings have been falling on the deaf eyes of Mankind's spirits ears; and mainly only what falls on the ears of his brain/intellect he hears. And with this no proper attention has ever been paid to finding out how things are and are also ordained to us to think about and investigate.

Now consider the following: “And God said unto them, be fruitful and multiply and replenish the earth, and subdue it: and have dominion over the fish of the sea and over the fowl of the air, and over every living thing that moveth upon the earth.” In these words lie the call for research and development which we have not properly heeded perhaps not understood.

Christ in his teaching, gave Mankind the ‘Sermon on the Mount’. In it among others he said, “seek and ye shall find”. He added knocking and asking. And since Jesus Christ brought the Truth from God the Father, His teachings must be fully all embracing. And so, the seeking, asking and knocking also point to research and development. And the words point to the fact that what we seek, ask for and knock already exist, but we must make the necessary effort by acquiring necessary effort to find what we seek. And no one can find if he does not seek. And in the process one should improve what already exist.

And he who seeks seriously shall always find something. But where is the necessary seeking Christ is referring to be found. Let us also add the following from Abd-ru-shin:² “Man should not believe in things he cannot grasp! He must try to understand them; for otherwise he opens wide the door to errors and with errors, the Truth (from God) is always debased...so we are to test and investigate”. And in another occasion He said, “The privilege of being endowed with the ability to think also brings man the duty to investigate.”

And as the Pharisee fought tenaciously against Christ and the Truth He (Christ) brought from God to us all as the only way to true knowledge, He said the following to them “Ye are of your father the devil, and the lust of your father yea will do. He was a murderer from the very beginning, and abode not in the Truth, because there is not Truth in him. When he speaketh a lie, he speaketh of his own; for he is a liar and the father of it.” All these are the raging evil forces and types we have produced which are now harming us all. Now consider wars, terrorism, militancy, violence and all else that lead in one way or the other to acts of murder, and how much is spent on these at the expense of beneficial research and development as well as acts of goodness in creative thinking and innovations which could smoothen our lives and living better than we are today. Are we even innately able to do as others have done for themselves? If no why?

Some Human Opinions

Consider all the Creeds Dogmas and Doctrines from various religious organisations. I am more familiar with the Christian Religion: Sooner or later all untruth will collapse for it will never stand the test of time or Eternity. Hence reflect on “Christ has no earthly father, salvation through the Blood of Christ, the meaning of the Virgin Birth of Christ; and the resurrection of Christ's physical Body from the Grave. Can

all these be true from all that we know from science engineering and technology (SET). Here we must investigate so as to know the Truth.

And if this triad of dogmas, is not true, is it not time to review the situation in the light of the ever increasing pressure of the Truth from God? And if later (and may be very soon) this triad is a lie, how shameful then that the proponents of this triad have been worshipping Lucifer for he alone passed these terrible lies to the various organisation which took up the lies and made the followers or peoples to believe them without questioning the truth behind them all. Even biologists of great eminence believed these doctrines. How much resources have been spent to maintain these dogmas for over two thousand years; and how much longer will they be maintained and in every case, it is the shackling of the free spirit of man, and its enslavement within the body. And if there has been research and development (R&D) of the true world around us, both the visible and the invisible parts of the World (or the Universe), wont things be different today? I wonder. And indeed there will be more resources and funds for carrying out R&D projects which should lead to a better life for us all.

In these opening remarks, I have tried to show that, in totality very little has been done by ways of true research so as to find out more and more of how things are or indeed ordained by whoever is responsible for all that are ordained working faultlessly in the Mighty Universe in which we have our being. However, the Almighty Himself is responsible for the Truth and the perfection that there is.

The Leadership Problem

It is not in my nature to blame nor condemn, but rather to ask leaders to review the direction leadership is going. In many things the people follow the examples their leaders are showing. If therefore leadership follows mainly the way of materialism, people themselves (with a few exceptions) will follow the ways of the leaders. For example, Jesus Christ is perhaps the greatest leader of all times on the Path of the Truth from God. His Goal is to serve the Will of God as He taught the Truth from God (Heaven) to all Mankind and He proclaimed “I do the Will of My Father Who sent me.” With this as the Goal of His Work and after overcoming the temptations of Lucifer, He set out to teach the Truth unerringly. And with this, He lived and showed how to live an exemplary life; and did direct that we should know, and investigate the Earth because our bodies come from it. Therein lies the eternal directive on farming and in all agriculture. Therein lies an eternal leadership system Christ introduced.

Science and Technology

Science, Engineering and Technology (SET) are concerned with knowing the earth in every way possible and harnessing the resources for the betterment of man’s life on earth. We can see how the research and development works of others have helped us in the obvious development we can see on the ground around us: roads, water supply, electricity, telecommunication and all else that makes living worth it.

However water and electricity have not been regularly available to the populace. Tarek Khalil⁵ in his book, “The Management of Technology” defined technology as “All knowledge, the products, processes, tools, methods, and systems employed in the creation (manufacture) of goods or in providing (or rendering of) services. In simple terms technology is the way we do things. It is the means by which we accomplish objectives. Technology is the practical implementation of knowledge; a means of aiding human endeavour”.

Technology is today extremely effective in producing changes and the speed with which the world (earth) is changing. The engineer as the translator of science (after the pure scientist has done his work) should play a decisive role in determining the future course of human existence. He has the training and responsibility for which his technological competence uniquely qualifies him so as to discharge that responsibility placed on him by the society. The engineer, and to some extent the scientist must be prepared to deal with the ever increasing problems of far broader scope than those solved by the predecessors, and so continuing R&D in knowledge advancement will be necessary always. How then can this continuing R&D be maintained if funding is grossly inadequate; or if funds were for a change provided are wasted away by scrupulous researchers as also does happen from time to time.

Consider the annual ritual of the rice “armado” from which most of us benefit. Does it really benefit us as a nation or, it is actually anti-beneficial when viewed in the context of nations. Some of the rice comes from

Thailand and by so doing employment for more Thailand people. In addition we Nigerians greatly improve their foreign exchange earnings; and at the same time, we weaken our currency.

It is often said that Nigeria can produce all the food (ordained by God for us) we need to eat. And so this include the rice we ought to produce it. By so doing, we produce enough rice for ourselves process and package and eventually, through mutual exchange of goods, guarantee improved employment for our youths (male and female) as well as earn increased foreign exchange. But because we have not invested enough in the rice production industry nor is there any purposeful leadership in this highly agricultural production aspect of our economy, we remain rice importers and not exporters. And has there been enough R&D investment in rice production research, both from within and from without? I wonder.

The National Problem

There is this Law of Life which states: “Whatever a man sows, that will he reap many time over.” It can also be put as “As a man thinketh so he is” in thoughts words and deeds. If you apply these two sentences which have been with us from all eternity, we can deduce from them why a people is what they are from generation to generation. For example, if a people follow the path of production, all the benefits of production will flow to them. On the other hand, if a people in thoughts follow the paths of lust and negative passion, greed, hatred, envy, and above all selfishness as well as corruption; and these dominate the minds of men and women in various positions of authority, then very little will be left in thoughts to plan what is most necessary for continuing development; And research and development are parts of the good paths of a nation, which is usually guided by love and knowledge.

What a people can do at any point in life is related to their total development effort in all aspects of total character development. Fundamental to everything is the correctness and relevance of all aspect of education. If most aspects of education are not relevant to national development and if national development plans are not what they should be, in the employment of the youths, the goals of wealth creation will elude the nation. I think we are in this situation to some extent. All these are happening because very little attention is paid to socio-economic development; because today’s world is based materially on the power of technology n the control of human environment. Whether for the good or for the evil which some times comes from technological applications such as war, production of dangerous chemicals and much else. Once more investment in research is grossly inadequate.

Lawfulness Energy and Knowledge (LEK)

We live in a world governed by Lawfulness Energy and Knowledge (LEK). It is absolutely impossible for a person to do anything without simultaneously using the necessary energy and knowledge; as well as what is done is lawfulness or not-lawful. This triad (LEK) will go a long way as a tool we can use to promote all R&D leading to gaining the much needed more knowledge through research using experience gained from knowledge to harness more energy and ensuring that what is done is lawful. Indeed every action must simultaneously take this triad of LEK into account. It will require a whole book for deeper understanding of the concept; for it goes beyond the frontiers of human intellect activities to realms of spiritual nature of man were taken into account in all triad of LEK the whole investment in science, engineering and technology will follow the pattern already well anchored in Nature. It is because it is said that Nature is our best teacher and Nature is always right.

If only the understanding of Nature were to form the basis of R&D investment, outstanding results, benefits, peace and true joy would have been attained on earth by now (or today) and therefore in our country. To invest on material things alone without taking into account the spiritual/eternal nature of man cannot make the complete human being, and this has been the case almost throughout the ages.

At this point in time in the presentation, we must recognise that the Lawfulness in the whole Creation of God is adamant, inexorable and unchanging from age to age and from generation to generation. Three Basic Laws govern all that manifest after every happening in the Mighty Universe – visibly or invisibly. Let us consider only one of the three, that is: “Whatever a man sows, that shall he reap many time over”. Put differently He who does not sow will not reap either. In these two sentences lie all the rewards, punishments and retributions in the Justice of God and the absolute mechanisms of total and manifestations in the workings of the Universe. If therefore we do not invest adequately in R&D, how else can we

advance knowledge necessary for the continuing progress and advancement of our nation and people. Can we really progress if we continue to break God's Laws with impurity. God's Laws are there, visibly and invisibly for us to know and use as the guardians of the eternal order of the Will of God. When it is said that, we human beings are to improve what already exists and further them; this we can do through research, development, thinking, creating, innovations and in all types of investigations. It also means we should awaken our intuition within us, to which each can listen to (as the still small voice within). And only in these various ways and applications we can develop ourselves from within; and with time, greater and ever greater scientists, engineers and technologists will arise and so the nation will blossom forth to ever higher levels of life that will eventually be pleasing to God, our Creator.

Take a trip to any part of the country. You will see the sufferings we have imposed on ourselves through not investigating and not wanting to do things according to God's Will. This, not wanting to live according to the Will of God is indeed, the fundamental sin/evil or wrong doings of men, dating back to the fall of man (FOM). We can achieve nothing if we do not want to know through appropriate education and research as well as the necessary investment so as to forever advance the frontiers of knowledge.

Since independence (1960) much has happened but much more could have happened if we have paid more attention to R&D, LEK as well as appropriately invested in them over the years. The fundamental requirement for all life and living absolutely depends on knowing the Laws and living according to Law. Joshua (1¹⁻⁹) was permitted to recognise this thousands of year ago: "This Book of the Law" shall not depart out of thy mouth..." That is the knowledge we can say "The Knowledge of the Truth" is fundamental to the good life guided by knowledge and love.

Agriculture – Nature Research

What is being said in this paper is basically that we have not invested enough in various aspects of science, engineering and technology through research and development so as to derive the best benefits from R&D work done. One can mention many research organisations such as NIFOR, Benin City, ITTA, Ibadan, Root crops research at Umudike, etc. The disappearance of the groundnut pyramids, and rice fields absence and the general lack of large oil palm farms as well as rubber farms are obvious examples of the gross inadequacy of research nationwide. Only the Federal Government has research setups. I am not aware of research establishments by the various States: and certainly none by the Local Government units. How can we know how to do things in thoughts, words and deeds, if we do not have the necessary investment in research so as to gain experience through investigating and executing projects for ourselves and by ourselves?

There is a serious and utterly urgent need to accelerate the research and development creative and innovative work in all aspects of Science, Agriculture and Technology (SAT). Globally, technology (like a body of knowledge) drives any nation's economy; and this situation must now be of concern for all leaders at various sectors of leadership that can be considered for our well being now and in the years ahead. And at this point in time we ought to look towards Nature; for Nature is our best teacher⁶ in all aspects of SAT development.

Trade Indexes

Let us give an example as a result of the general inadequacy of investment in research and the why of the low value of our currency. Obviously it is basically a people or a race's problem who are yet to totally awake to the reality of the true life and living.

Consider that a trade balance of a nation represents the difference between the total value of merchandise goods (ΣV_{ge}) and the Services (V_{ge}) exported by that country and the total value of merchandise goods (ΣV_{gi}) and services (ΣV_{si}) imported. The trade balance equation is:

$$V_{gtd} = (\Sigma V_{ge} - \Sigma V_{gi}) + (\Sigma V_{se} - \Sigma V_{si}) \quad (1)$$

The sign Σ is the summation of all the goods/services in the export/import global trade of the nation concerned. The trade balance is an index of the relative competitiveness of a country's industrial productivity and services organisations. The trade balance is either positive in which case the country has a

competitive edge to other countries; and if it is a trade deficit it causes inflation exchange rates and automatically means a weak currency for the nation as we are today. No one thinks about curing this disease.

Every person in a country plays a part or makes a contribution to this balance of trade equation. Any one taking more than he/she puts into the national economy is therefore causing inflation either to the goods' part of the equation or the services' part of the equation. As the value of the nation's currency declines due to the trade deficit, it becomes more expensive for the people to buy imported products. And on the other hand their exported goods become cheaper to other countries' markets which trade balance is positive. This is the situation we have found and put ourselves as a result of perhaps our spiritual indolence and intellectual laziness and love of ease for other people's products for even those goods we can produce, we end up importing them.

Investment Indexes & a Nation's Competitiveness

Competitiveness is the process by which an entity (or a nation) strives to outperform another or better put maintain a competitive role in the global trade. But no country can stay competitive if there is no adequate investment; in which case the economy cannot operate successfully and that country's competitive edge will be poor. To be competitive in world trade the nation's productivity must improve. And productivity is the efficiency with which goods and services are produced and provided. It is determined by the nation's total workforces, technological creativity and innovation using the resources of Nature at our disposal, the quality of the plant and equipment used, and the effectiveness with which these factors of production are used. At every step, the relative investment must be right in every way. Here the human capital, plant and equipment must be adequate.

"Investment is the fundamental building block of current and future economic activities of a nation and a people, and it is therefore, at the base of the nation's competitive pyramid, Fig. 1.



Fig.1: The Competitive Pyramid

Source: "Management of Technology": Khalil (2000).

Investment is also the fundamental determinant of national competitiveness. The definition includes soft assets, such as public and private sector spending on R&D, SAT, public sector expenditure on education.

The Science, Agriculture and Technology productivity of a nation is the ratio of output and input in each of the sectors mentioned above. It means or reflects the efficiency of an operation. In this respect, the common index used to track or measure productivity is the output per worker – hour input, shown in Fig. 2. We are not able to place the Nigerian case for the period under consideration. However the message for us in the diagram is clear. It is the measure of a peoples output of the productivity if the relative investments are adequate. The Japanese case is outstandingly clear, the result of a people's productivity.

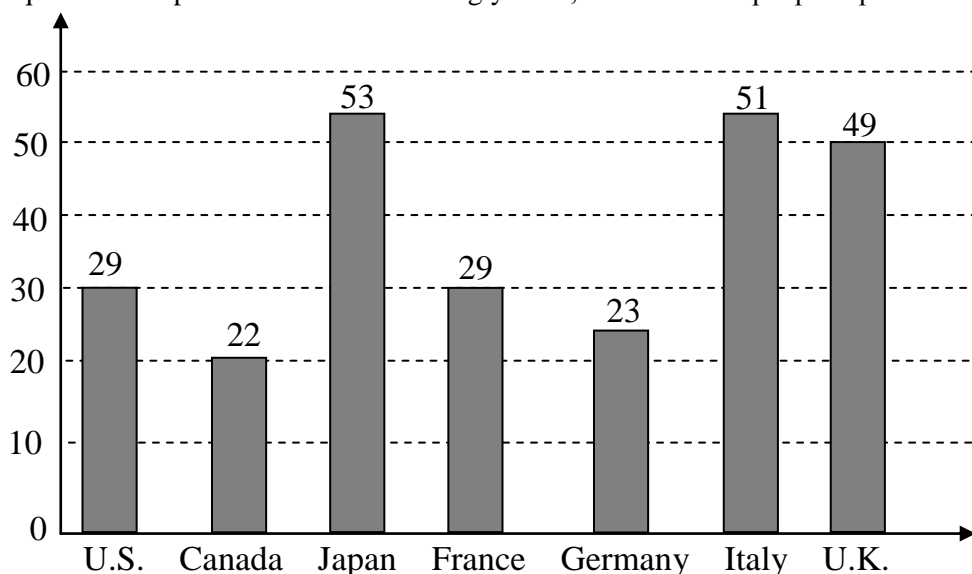


Fig.2: Long-Term Real Growth in Manufacturing Productivity, 1983-1993

For this chart, manufacturing productivity growth is based on output per manufacturing hour.

Source: Council on Competitiveness, 1995 Note: The Nigerian case is not shown

Investment Indices³ (Indexes)

Investment in R&D, Science, Agriculture and Technology (SAT), plant and equipment (P&E) and appropriate education provides a base for long term economic growth. It is therefore most important to track these indicators and to cry out or sound a clarion cry or even jingle the alarm when they take a wrong turn. Clearly they have taken a wrong turn and the time is over due to the fact that we should have cried out long ago. We must not give up, but double our effort to see to the adequacy of investments in R&D, SAT and all that is necessary to become competitive in the every expanding global market for which we are a part.

Solar Dry Research Development

It is important to briefly mention an ongoing work for drying perishable food items. The creative and innovative design comes from the internet⁵. The basic unit is as shown in figure 3. The design consists of one drying tray, the solar heat absorber and radiator. The heat flow system in the container is clearly shown. It is a simple heat and mass flow system.

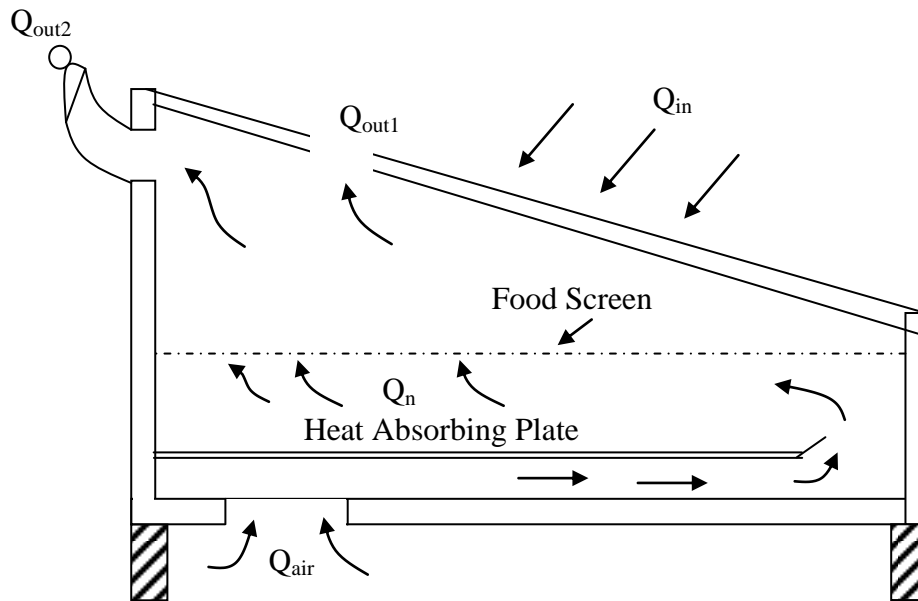


Fig.3: Experimental Solar Dryer

The Heat Flow Equation is:

$$Q_{in} + Q_{air} = Q_{out1} + Q_{out2} \quad (1)$$

The heat balance equation is:

$$Q_{solar} + Q_{air} = Q_{out} \quad (2)$$

If it is assumed that there are no losses.

Q_{solar} is obviously dry heat passing through the glass shield, Q_{air} is the heat inlet from the bottom through the holes/vents which are screened and Q_{out} is the moisture contained heat which carried away the moisture/moist content of the materials being dried.

The method can be modified to include electric and charcoal options. With time, the work will include through innovative, creative and investigative approaches in finding solution to drying food items which otherwise are perishable and become wastes. If the items are properly dried they can be properly packaged and put on shelves for sale. The information available from those who have done some work on the subject is that solar dried products can last for more than one year and they store freshly dried and they also taste well. The reason for better taste will be a subject for further investigation.

This topic is just mentioned to show that investment and innovative thinking are simultaneously necessary for any R&D work to progress.

Conclusion

It is shown in this paper that investment in R&D as well as in SAT is grossly inadequate. Some basic philosophies of life considered to be aspects of the eternal Truth from our Creator were first presented. Later the subject moved on to show how the low investment in research and that will make our global competitiveness effective were mentioned and briefly explained.

It must now be repeatedly stressed that a nation's technological advancement is a major (or indeed the major) contributor to its economic prowess. Through R&D, the Management of Technology (MOT) will play a major role in creating, innovating and maintaining a competitive role in the global market place. MOT activities may be undertaken at the national/international or macro, level or at the firm micro level. On the whole countries must be able through R&D to;

- (a) Create an economic growth policy noting the fact that technological policy is a major contributor to economic growth.

- (b) The necessary infrastructure permitting the support of technological enterprises and the facilitation of commerce industry and trade. Planning the human capital⁷ (or resource) development must also be an integral part of any technology development strategy starting at rural village levels.
- (c) It is absolutely necessary to encourage cooperation between government; industry and education and research institutions.
- (d) We should energise and support technological creativity and innovation and develop plants to enhance creativity and support R&D activities through continuing and ever increasing investment.
- (e) Promulgation of necessary but unburden some legislations and regulations measures to protect the environment and strengthen social structure.

A nation's competitiveness depends on its successful exploitation of raw materials, labour, transportation, the resources of financial and human capital. These factors are always important, but further aided by the advantage of the explosion of today's knowledge to always create advanced technology that will help and maintain a competitive edge.

There are changing world conditions as well as the changing environment of business for the global market. Therefore the competitive advantage is increasingly dependent on our talents, gifts and abilities' and skills development in managing technology and technology enterprise.

Therefore a nation's technical enterprise depends upon the following factors (Khalil, 2000).

- The strength of the national research enterprise
- The quality of technical education
- The presence of a large pool of technical talents.
- The strength of information technology infrastructure.
- The ability to cultivate individual creativity and initiative.
- Synergy between basic research and downstream technical activities such as design and production capabilities.
- The scale of domestic markets and the openness of global markets as engines for innovation and its commercialization.
- The ability to continually modernize plant and equipment in private industry, and the commitment to do so.
- Collaboration between industries and universities and the government.
- National savings and the level of investment in industrial modernization.
- National policy supporting initiatives to enhance adoption, adaptation, and diffusion of technology and related know-how.
- The development of the necessary human, physical, financial, regulatory, and institutional infrastructures to attract individuals, companies, and institutional entities, regardless of national origin, to invest in and conduct technical activities within the boundaries of the country. This ensures the long-term wealth-generating capacity of the economy.
- Public support of generic and domestically developed technologies.

Entrepreneurship and Investment

Entrepreneurship is the necessary philosophy for starting and running a business project in such a way as to bring out better well being to people and indeed the whole environment. In all these, R&D leading to one type of investment is inevitably necessary. Entrepreneurs by their very nature are people of vision, courage, initiative, commitment, persistence independent thinking drive to succeed and ambition. Some entrepreneurs have an appreciation for a particular technology branch, good motivational skills and a commanding personality.

Notable entrepreneurs who have made names for themselves are Bill Gates and Konosuke Matsushita, often called the most remarkable 20th Century Entrepreneur. It is known that investment is the foundation

for all entrepreneurial success flowing from necessary research and development work. Much can be said under this section, it will always point to taking risks in investment of one type or the other and leading to one type of economic growth or the other. Once more we have not made much progress in this either. For, entrepreneurship is known to have pushed fast a nation's wealth creation, ventures and jobs expansion than big industries have done.

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INVESTING IN PHYSICS IN 21ST CENTURY NIGERIA: PROBLEMS AND STRATEGIES FOR IMPROVEMENT

P.E. Asarhasa and E.E. Enukpere

Physics Department, College of Education, Warri, Nigeria.

E-mail: parhasa_8@yahoo.com

ABSTRACT

Science and technology the saying goes, “accelerates progress”. Hence the concern of every society especially the developing ones in the 21st Century is to advance both scientifically and technologically. The impetus for this is substantial investment in science. However, the quality of funding for physics programmes leaves much to be desired. Consequently, the authors re-examines the importance of physics in National Development. Constraints preventing physics activities from becoming comprehensive in its economic impact in Nigeria were highlighted and ways for improving support for physics practice in this era of globalization were also discussed.

INTRODUCTION

The scientific enterprises of the developed nations are traceable to their long-time recognition of the need to support capacity in science in order to solve inevitable problems and develop informed policies (Parks, 1996). Bruce (2004) hinted that no nation can now afford to be without access to a credible, independent science and technology (S & T) research capacity as these have the potential to propel such nation into the orbit of scientifically and technologically advance nation of the world.

In the struggle to get developed, nations especially developing ones face a broad array of challenges that require the application of up-to-date scientific knowledge and technology to resolve. Such challenges include stimulating economic growth, mitigating environmental problems, safely adopting beneficial new technologies and quickly responding to sudden outbreaks of new diseases. Nigeria as a developing country is not isolated from these challenges.

That Nigeria is presently struggling with deep and resistant socio-economic difficulties is a fact that required no proof. Nigeria's economic woes are attributable to her low funding of science activities on one hand and little or no patronage of such activities by the private sector on the other hand. This state of affairs is not wholesome for rapid growth and development of the nation. We believed that these challenges are surmountable through substantial investment in physics.

It is in a bid to reverse this that these authors re-examine the importance of physics in national development; pointing out ways for improving support for science activities in this era of globalization thereby accelerating the nation's progress in the area of economic growth and development.

PHYSICS KEY ROLE IN NATIONAL DEVELOPMENT

Physics is a natural science, it is the study of matter and its motion through spacetime and all that derives from these such as energy and force. Maxwell (1878), More broadly, it is the general analysis of nature, conducted in order to understand how the world and universe behave Feynman(1963). It is one of the oldest academic disciplines, perhaps the oldest through its inclusion of astronomy.

Physics is both significant and influential, in part because new ideas in Physics often translated into new technology, but also because new ideas in Physics often resonate with the other sciences, Mathematics and Philosophy. Roserberg (2006), Wikipedia (2008) Physics as a discipline is the bedrock of modern scientific technologies.

For example, advances in the understanding of electromagnetism or nuclear Physics led directly to the development of new products which have dramatically transformed modern-day society (e.g television, computer and domestic appliances). Advances in thermodynamic led to the development of motorized transport and advances in mechanics inspired the development of calculus.

Applied Physics is a general term for Physics research which is intended for a practical use. Applied Physicist are using Physics or conducting Physics research with the aim of developing new technology or solving a problem. For example people working on accelerator Physics might seek to build better particle detector for research in theoretical Physics Young (2004).

Further more, Physics is used heavily in engineering, for example, statics, a sub-field of mechanism, is used in the building of bridges and other structures. The understanding and use of acoustics results in better concert halls; similarly, the use of optic creates better optical devices. An understanding of Physics makes for more realistic flight simulators, video games and movies and is often critical in forensic investigations.

With the standard consensus that the law of Physic are universal and do not change with time Physics can be used to study things that would mainly be mired in uncertainty. For example in the study of the origin of the Earth, one can reasonably model Earth's mass, temperature and rate of rotation, over time.

It also allows for simulations in engineering which drastically speed up the development of a new technology.

Also, there are whole fields of apparently independent discipline which are of Physics in origin e.g. electrical engineering, electronics and the World Wide Web (www) which was developed at CERN for the use of Physicists which have developed into totally independent fields.

No wonder Physics as a discipline is regarded as the bed rock of modern Scientific technology. A nation's development is measured in terms of her industrialization. The industrialized nations are the world most technologically advance nations and have grouped themselves into G - 8. These nations are characterized by a high output of industrial goods and their economies are very buoyant. In each case, they all possess nuclear power not just for energy production but also for war fare.

Modern technology rests on science and of particular interest in Physics.

From the above, it becomes obvious that Physics and indeed activities of Physicists are vital in national development.

Firstly, they facilitate the expansion of the local industries through the development of fundamental technologies which may give birth to new industries and materials. For instance, various principles of physics are used by geophysicists, reservoir engineers and petrophysicists in the exploration and exploitation and production of oil and gas in the oil industry while the same is true in the extraction and processing of solid minerals. All of these, you will agree with me has constitute the mainstay of the Nigeria economy.

Secondly, physics research discoveries have a complex and multidimensional effect on national development. Sam (1991) Physics research findings has revolutionized the economy of most nations of the world and helps them tremendously in raising the general level of scientific awareness of people and draws young minds towards careers in physical science and associated area of technology. Countries that see science as an essential part of their future wealth and well-being actively encourage research in physics. Ubachukwu and Okeke (2001).

Nigeria search for a stable and cleaner energy supply (i.e solar, nuclear energy etc) has not yielded the expected result because the fundamental technology used in harnessing such resources and which are physics research based has not been properly developed and supported.

Also, the country's hope of joining the space club has remained a mirage for the same reasons. Needless to states here that the technologies associated with solar energy, nuclear physics and space physics determine the economic and military power of a nation.

Countries without these potentials are classified as an underdeveloped. According to Ubachukwu et al (2001) development does not mean the ability to purchase ready – made products of space technology, such as satellites, cellular phones, aeroplanes and hiring of drilling rigs for deep water exploration of oil and gas. Rather, development is the unfolding of peoples imaginations and liberation to begin to assert authority and self-reliance in carrying out human activities.

Therefore, that few countries (i.e the advanced nations are monopolizing these scientific activities pose a real and serious danger to Nigeria. The multinational oil companies that are neck-deep in on-shore and off-shore operation in this country and all other such companies operating in Nigeria have their research centres / laboratories located far away in their home countries making Nigeria a dumping ground for the products of research carried out abroad.

This is no doubt has been seriously affecting the Nigeria Content Drive (NDC) of the government for the oil and gas industry. Mayoma (2008).

From the foregoing, it becomes imperative that establishment of and effective running of physics research institutes and centres is an essential component in national development. For they create confidence in research and encourages scientific culture and create jobs for scientists bringing a spiral progressive movement for the country's development as a whole.

CONSTRAINTS TO PHYSICS ACTIVITIES IN NIGERIA

Nigeria as a nation has always realized the primacy of Physics programme in its national developmental effort. Hence to ensure a good head start all science disciplines of the country universities were encourage to undertake scientific research activities. Fafunwa (2004). Also national research institutes were established to provide solutions to developmental problems.

However, recent reports have shown that science programmes across the country both in institutions and industries are on the decline. Adegoke (2008) decried the alleged low state of research - oriented programmes in the country, noting that the development if left unreversed would continually hinder the economic growth of the nation.

According to Okonofua (2008) a numbers of constraints are preventing Physics activities from becoming comprehensive in their economics impact in the country. Among these are:

- 1. INADEQUATE FUNDING:** One of the major constraints to science and technology development in Nigeria is inadequate funding. Nigeria's total expenditure on scientific programmes is about 0.2% GDP which is very low when compared with those of the advanced nations like India, 1.2%, Brasil, 0.97% China 0.69%. 0.2% is far low from the minimum target of 1.0% GNP recommended for developing country in the world plan of action by the United Nation's for 1980. This problem of inadequate finding is sometimes compounded by some scrupulus administrators of tertiary institutions, research centres and researchers who wasted away such funds that are meant for a change.
- 2. PERCEPTION OF GOVERNMENT AND PRIVATE SECTOR TOWARDS INVESTMENT IN SCIENCE:** Another factor inhibiting scientific activities in developing countries especially Nigerian is Government and private sector's perception of these activities. Investments in scientific research are perceived as a time consuming, wasteful and costly activities by both the Government and the private sector. In the case of Nigeria Blue-ships companies and Commercial Banks are so neck-deep in investing in oil business due to quick returns of the so-called petrol-dollar. It must be emphasized here and now that developing nations should

understand the fact that perceiving investment in scientific activity as a time consuming, wasteful and costly activity will bring further limitation on their economic growth and development.

3. **SHORTSIGHTEDNESS OF POLICY MAKERS:** Genuine effort by our fully trained scientists to make their contributions to national development have over the years has been frustrated by policy makers. As Ukoli (1985) asserted, the preoccupation of Governments and the general public with the motion that scientific research must be geared towards solving immediate problems of local relevant is one of the greatest stumbling blocks to meaningful investment in Physics programme and hence to creativity amongst Nigeria scientists.

The universities and other research institutes are thus starved of funds for basic research, a situation that is compounded by understaffing in term of academic and technical personnel, and deficiencies in basic amenities like water, electricity, library and ill-equipment laboratory, communication facilities among other factors. Most of the laboratory due to paucity of fund are ill-equipped and ill-maintained. According to Ukoli (1985), under this prevailing conditions, it is virtually impossible to expect a meaningful research that would lead to innovation and invention that could attract investment and support. Pertaining to the issue of research relevant to immediate local needs, these authors are of the opinion that research findings which are not relevant to our immediate needs do not necessarily make them useless. Hardly does a society exist where all research findings are relevant to immediate society. The future is relevant and so is research into its problems and needs.

Science and technology budgets receive little or no patronage from the private sector and instead depend on the national treasury Asgar (2005). And considering the meager 0.2% GDP expenditure from national budget, coupled with heavy Government bureaucracies all combined to wind-up cultivating whatever science and technology is fashionable in the developed countries, waiting indefinitely for the time when such competence would trigger development in developing countries like Nigeria.

These constraints and many more have created an unfavourable environment in which scientific enterprises (physics in particular) will be expected to thrive and reach its peak.

STRATEGIES FOR IMPROVING INVESTMENT IN PHYSICS

Science and Physics in particular has a strong base from which to expand and to deliver excellence to be central to the developmental agenda of the country, Nigeria. But the past 20 years have seen a growing realization that current support for Physics activities are inadequate and may continue to make our current vision 20-2020 unrealizable. Unless we start to make real progress towards improving support for scientific activities we will all face a future that is less certain in economic growth and less secure in National development.

The following key points may be considered as recommendation.

- 1) The need to aligned research with national priorities to increase impact and perceived importance of research activities.
- 2) Another key solution is the establishment of centre of excellence and centralized research laboratories in selected universities with up-to-date laboratory analytical equipment. Such centre would attract better funding and consultancies from the private sector. Additionally, the improvement of the basic research capabilities of universities would also enhance the Nigeria Content Development (NCD) drive of the Government for the oil and gas industry.
- 3) A more realistic models that a developing countries should follow to attract investment in Physics is the practical needs model that is, demand – influence supply, namely, the type of pure investment that is done for example, after solid-state devices such as transistors made possible the expansion of switch boarding in telephone services, industrial laboratories such as Bell Laboratories financed solid state physics.

- 4) One other strategy is that reflects local priorities and specifies available funding – in consultation with that countries science, engineering, medical and industrial communities. Each nation's strategy should include support for basic science, education, and training that will allow it to achieve local competence in selected areas of national priority. To this ends, these authors suggested that developing nation's commit a minimum of 1 percent to 1.5 percent of their GDP to S & T capacity – building and stress critical importance of using competitive merit reviews to allocate these resources.

CONCLUSION

In this paper, an attempt has been made to highlight the role of Physic in national development.

Constraints inhibiting Physics activities from becoming comprehensive in their economic impact in Nigeria in the 21st century were outlined and strategies for curtailing these problems suggested. Additionally it was emphasized that inadequate finding is a major constraints to quality Physics practice in Nigeria and it has a multiplier effects on major facilities such as computers, laboratories, libraries e.t.c. This, it is argued can only a throne frustration, apathy and unproductivity in scientific pursuits. Investment in Physics like any other science subject we must remember, is the fundamental determinant of a nation competitiveness.

Apart from providing a solid base for a nation's long-term economic growth, it also serves as the fundamental building block of present and future economic activities of a country and a people. Therefore, it becomes imperatives that adequate funding of science activities in the form of establishment of and effective running of Physics research institutes and canter is an essential component in national development in 21st century Nigeria.

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CHALLENGES AND PROSPECTS OF CONNECTING UNIVERSITIES IN DEVELOPING COUNTRIES TO THE INTERNET : THE DELSU EXPERIENCE

G.E Akpojotor¹ G. C. Omede² J. Ebasa³ and E. U. Omede⁴

¹Department of Physics Delta State University Abraka, Nigeria

²Institutes of Information and Communications Technology

³Department of Mathematics and Computer Science Delta State University, Abraka, Nigeria

⁴Bursary Department, Delta State University Abraka, Nigeria

Abstract

The world is now regarded as a Global village with the internet as the Town square. Therefore any information in such media as books, magazines, journals, data, etc that are available on the internet can be accessed from any part of the world that is connected to the World Wide Web. The implication is that the potential to abridge the information gap between the developed and developing countries is now available. We have observed in this study, however, that many of the universities in developing countries have not been able to take advantage of the Web. The challenges facing them are both general and local. Some of the general ones include inadequate power supply, overloading of bandwidth, lack of funds/cost, security issues, lack of information, communication and technology (ICT) expertise and the inadequacy of the knowledge of ICT by universities management. On the local scale, we have used the challenges at the Delta State University as a case study. We then consider the prospects and proffer solutions to the challenges both on general and local scales.

Introduction

The world is now regarded as a Global village with the Internet as the Town square. Therefore any information in such media as books, magazines, journals, data, etc that are available on the internet can be accessed from any part of the world that is connected to the World Wide Web. The implication is that the potential to abridge the information gap between the developed and developing countries is now available as the internet provides a theoretically borderless, potentially infinite space for the production and circulation of information (UNDP, 1999). This ever growing and improving technology of our time offers access to useful academic resources and services as well as provides medium for exchanges and collaborative interaction between individuals and their computers without regard for geographic limitation of space. However, it is not actually being effectively utilized in many universities in developing countries including Nigeria, due to some bottlenecks that impair effective connection to the web (Jagboro, 2003; Poda et al., 2006). In a survey conducted in two Nigerian universities in 2005 by one of us (Akpojotor and Ojobor, 2005), it was observed that only 21% of the 80 participants were then using the Internet to obtain information relevant for their research areas. This is very sad especially as they observed, by visiting the libraries of these institutions, that they were not stocked with current journals, magazines, books, periodicals, etc. The authors could not help wondering what kind of research the other academic staff (with about 50% of them aged between 25 – 44 years) that do not use the internet yet for the purpose of research, are engaged in. Thus they were compelled to remind these researchers the old humble mockery of isolated researchers in science and technology (S & T) in developing countries, “Einstein cannot be born in a jungle.”

The observation of poor usage of the internet in the two universities surveyed by Akpojotor and Ojobor (2005) gave an insight into the national scenario. It supported an earlier study (Adeya and Oyeyinka, 2002)

where it is observed that the level of internet access and connectivity in Nigeria is far below that of developed countries. In a broader study, a survey by the Association of African Universities in 1998 found that only 52 of the 232 academic and research institutions had full internet connectivity, the remaining 180 institutions had access that was deemed inadequate (Useem, 1999). This is not a good rating and prospect for our educational goals and aspirations. Internet access availability in universities is a necessity that should not be overlooked. Kamba (2009) saw the Internet as a developmental agent having impact in the research process and information dissemination. Thinking in line with these assertions, the Delta State University (DELSU) branch of the Academic Staff Union of University (ASUU) in partnership with the university management contacted an IT firm to provide internet services to its members. The cost of the facilities and installation is about 4.2 million Naira and the cost of the bandwidth was about 4 million Naira per month. The cost of running it for 24 hours was enormous because it was powered by a standby generator that was consuming about 50 litres/month of diesel. This consumption which was only at the Abraka campus, does not include the 10 hours of power provision by the university management to the university community whenever there is power failure from the National grid. Indeed, the DELSU ASUU experience of internet service provision to its members is a lucid example of the challenges faced by a Nigerian university in connecting to the internet. We shall therefore use it as a case study. Interestingly, the service is no longer available and the providers are seriously considering a number of issues that will enable them reactivate the service. We shall therefore proffer some probable solutions to surmount the present DELSU ASUU internet comatose. The solutions are expected to be applicable in other universities in developing nations as such they give possible prospect to connecting universities in this part of the World to the internet.

The plan of our presentation is the following. We will begin with the explanation of two important keywords: Internet and World Wide Web (www) before listing and analyzing the challenges and prospects of connecting the universities to the internet. Thereafter, we will conclude.

Internet

The Internet, sometimes called simply "the Net," is a worldwide system of computer networks - a network of networks in which users at any one computer can, if they have permission, get information from any other computer (and sometimes talk directly to users at other computers). It was conceived by the Advanced Research Projects Agency (ARPA) of the U.S. government in 1969 and was first known as the ARPANET. The original aim was to create a network that would allow users of a research computer at one university to be able to "talk to" users of research computers at other universities (Gray, 1999). A side benefit of ARPANET's design was that, because messages could be routed or rerouted in more than one direction, the network could continue to function even if parts of it were destroyed in the event of a military attack or other disaster. Internet this day is a public, cooperative, and self-sustaining facility accessible to hundreds of millions of people worldwide. However, the Internet's early goal for promotion of research appaers in higher education as a tool for the researchers, teaching staff and students to communicate and share project data has continued to improve. Today the .edu domain is still one of the largest contributors to the Internet (Poda et. Al, 2006).

Physically, the Internet uses a portion of the total resources of the currently existing public telecommunication networks. Technically, what distinguishes the Internet is its use of a set of protocols called TCP/IP (for Transmission Control Protocol/Internet Protocol). This can further be networked into a local area network (LAN) or a Wide Area Network (WAN) restricted within an organization. Therefore, a university will not only use the internet for access to other people worldwide, it can also restricted its use within its organization. This restricted internet operation which is known as intranet is purely for effective organization teamwork and collaboration as it enable members of an organization to shares files, work as a team, ease project execution, etc, via the internet. We have studied the intranet conflicts in Nigerian Universities somewhere else (Akpojotor et al., 2010).



Fig. 1 (colour online): The Internet

World Wide Web (www)

The World Wide Web (www) is a system of internet servers that support specially formatted documents in markup languages. A markup language is used to write the source code for a document that gets compiled into presentation form that can be stored in digital libraries (Landau, 2008). For the World Wide Web, the documents are formatted in a markup language called HTML (Hypertext Markup Language) which supports links to other documents, as well as graphics, audio and video files. This makes it possible for one to move from one document to another and from one site to another by simply clicking on the hot spots as well as instant cross-referencing (Wilkie, 2002; Akpojotor et al., 2005)) and thereby have created an emerging new 'one stop shop' pattern of reading activity. Sometimes there are buttons, images, or portions of images that are "clickable" to view or animate the. If one move the pointer over a spot on a Web site and the pointer changes into a hand, this indicates that one can click and be transferred to another site.

Using the Web, we have access to millions of pages of information. Web browsing is done with a Web browser, the most popular of which are Microsoft Internet Explorer and Netscape Navigator. There is also the open source firefox. The appearance of a particular Web site may vary slightly depending on the browser you use. Also, later versions of a particular browser are able to render more "bells and whistles" such as animation, virtual reality, sound, and music files, than earlier versions.

Challenges

Challenges in this context of the presentation here simply mean those obstacles or factors that hinder the success of internet connectivity in Nigeria universities. This is one of the main focus of this paper. The challenges include though not exclusive

- ☐ Universities Management knowledge of ICT
- ☐ Lack of ICT Expertise
- ☐ Funding
- ☐ Bandwidth and its management
- ☐ Inadequate power supply
- ☐ Security Issues

☐ **Universities Management knowledge of ICT**

The importance of ICT to the Nigerian university system has long been recognized. This is why the National University Commission has made it compulsory for university to have an ICT unit that must be coordinated/managed by a senior IT officer. However, the implementation of this directive has been far from being effective because many of the universities' administrators lack sound knowledge of ICT and its importance in academic environment. Therefore their priorities are placed in other areas different from not ICT. This lack of knowledge can be viewed at various levels:

- At the formation level: this is the level to consider what should constitute the ICT requirements of the university. It is at this level the decision on whether to use it for internet general purpose, intranet, e-libraries, etc, has to be made. Implicitly, it is at this level also that the targeted users

have to be earmarked as well as the cost. Furthermore, the possibility of future expansion should also be considered at this level.

- **Implementation level:** this is the level to consider how, when and who to implement the required ICT in the university. The starting point is members of the university community to be involved in the implementation. As stated above, the NUC directive is that there must be an ICT unit to be coordinated by an IT expert. The implication is that the leadership of this unit must be given to experts and not been seen as mere academic political appointment.

The summary of this challenge is that many universities' managements lack understanding of ICT and therefore cannot formulate appropriate ICT policy in their universities. Further, this lack of understanding of ICT often results in not adhering strictly to the standard ICT policy in execution of ICT projects. The case is even worse when there are management staff who myopically have resolved that they cannot or do not have much interest to improve on their poor ICT level and therefore belong to the class of information want-nots (Akpojotor and Ojobor, 2005).

❑ **Lack of ICT Expertise**

Closely related to the challenge of universities' administrators knowledge of ICT is lack of ICT experts in our universities and we shall consider this factor as follows:

Inexperienced ICT personnel in Nigeria Universities.

Internet connection is a big project which should not be left in the hands of inexperience persons to fondle not only because of the financial involvement but also because of its sensitivity and relevance. However, it has been difficult to employ and retain professionally qualified IT personnel who should give sense of direction for ICT development in our universities. Therefore, most of the ICT personnel in our universities are paper qualified and have little or no practical knowledge of how to manage this project. This problem is not helped by lack of in-service training of ICT Staff. ICT in general, is a very dynamic field and therefore requires the regular in-service training of the practitioners to catch up with new developments through training workshops, seminars, etc.

Use of External bodies in Executing ICT projects.

Providing internet facilities is a major contract in the university and due process requires that it should be properly advertised for interested providers to tender their bids for the selection by the appropriate university committee and only the most suitable of them should be awarded the contract. Needless to state, this contraction is at the implementation level. Thus the achievement and maintainability of the university policy on ICT is being compromised not only because these external bodies such as the contractors lack knowledge of the inner workings of the university but also because the primary interest of the contractors is making their money. This was the case of DELSU ASUU internet. The first six months of subscription by an external body was about 36 million Naira which is a far cry from the about about 1.8 million Naira per month totaling 10.8 million Naira for 6 months paid for the same period when one of us took over the management of the facility. Thus the challenge here in having effective internet connection in Nigeria universities is lack of demarcation in commitment to awarding ICT projects and other projects.

❑ **Funding**

Computer Networking and hooking to internet which involves purchasing of internet facilities, contacting Internet Service Provider (ISP) and payment of monthly subscription fees could be very expensive. As stated in the introduction, the cost of the facilities and installation of the DELSU ASUU internet facilities is about 4.2 million Naira and another 2.5 million Naira when the first facilities packed up. The cost of running it for 24 hours was enormous because it was powered by a standby generator outside the hour the university was providing power to the university community in the absence of power from the Natioanl grid. The plan was that the academic staff would pay monthly subscriptions of N2,500.00 every month for a given period so that part of this amount will be used to pay back loan from the university for the cost of

the facilities and installation and the remaining part for the monthly subscription, maintenance and other running costs. One immediate problem is that some members do not have personal computers/laptops and consequently viewed the monthly subscriptions which were being deducted at source as extortion. On the contrary, there was a general consensus that the provision of the internet facilities should be the responsibility of the university management as directed by the NUC. Thus the issue of funding was one of the major challenges to the DELSU ASUU internet service and it is the same in many other universities in the developing world since most of these universities are not adequately funded, so that they try to use the fund they receive for other purposes which might be termed the priorities. It is pertinent to point out that the costs of Internet connectivity in developing countries especially in Africa can be hundreds of times higher than those in Europe or the United States of America. For “free” information on the Internet, institutions in developing countries must often buy larger-capacity connections than they can realistically afford. For example, some universities in Africa are spending as much as the equivalent of 20 full-time faculty salaries for a 2-megabit Internet connection that is then distributed to 500 to 600 computers, resulting in a costly and painfully slow connection for everyone (World Bank, 2006).

□ **Bandwidth and its management**

Bandwidth simply means the data transfer capacity of a transmission medium. In digital communications, the transfer capacity is expressed in bits per second (bps), kilobits per second (Kbps) or megabits per second (Mbps). For example, Ethernet accommodates a bandwidth of 10,000,000 bps or 10 Mbps. In analog communications, bandwidth is the difference between the highest and lowest frequencies in a specific range. For example, an analog telephone line accommodates a bandwidth of 3,000 hertz (Hz), the difference between the lowest (300 Hz) and highest (3,300 Hz) frequencies that it can carry. The Nigerian bandwidth for internet communication is generally small. Thus inappropriate management of bandwidth of data transmission media use in internet connection is another challenge that is faced in connecting Nigeria universities. This can be viewed as follows:

□ **Inadequate bandwidth size**

The bandwidth paid for most of the time is not enough for the systems to be effectively connected to the network, thereby causing traffic jam in data transfers, some packets must wait for others to move, etc. This manifest as sluggish network in which browsing becomes hectic and frustrating. The was another challenge of the DELSU ASUU internet facility: the size available to users was too small for effective performance for its members. Therefore, there were sporadic incidence of congestion especially during the day and this was frustrating.

□ **Choking of Bandwidth**

Another challenge of bandwidth management is lack of System bandwidth control which is the inability to assign bandwidth size to individual computer on the network. This often results in bandwidth overloading which in turn manifest as sluggish network.

□ **Inadequate power supply**

Power supply is one very serious problem in connecting universities in Nigeria to internet; the internet facilities are electronics and cannot work without electric power. The condition of power supply in Nigeria is not actually reliable for systems that need to be continually on. The problems in form of constant power outages and unstable electricity with power surges can distort the functionality of this equipment making it difficult for our universities to remain hooked up without interruption. Imagine a situation where the modem and other equipment were powered on, suddenly there was a power outage, putting off the equipment, only for the power to come back probably after some seconds, minutes or even hours, then one found the modem starts malfunctioning or some of its components damaged.

To protect against this and other hazards, a UPS, Inverters and a surge protect for modem and other internet facilities should be readily available. There should also be standby generator or even solar system for big organizations to support the public power supply. However, the cost of purchasing, maintaining and running standby generators by these universities is enormous. For example, it cost a mean value of N7.26

million to purchase 66, 000 litres/month of diesel at official price of N110 to run the Delta State University standby generators at Abraka just for about 10 hours daily and this include 6.00 a.m. – 8.00 a.m, 12.00p.m – 2.00 p.m. and 6.00p.m. – 12.00 a.m.

□ **Hardware and software decision**

The internet facilities comprise of hardware and software and they form the core of the implementation of internet connection. Usually, the decision on what hardware and software to install depends on the planned usage and coverage. In general, some of the guiding principles should be based on important issues such as speed, security and maintainability and upgradation.

Speed: The speed of the internet cannot be over emphasized. Though the bandwidth management plays an important role in the speed, the hardware and software installed are also vital. We do not wish to discuss the details of the various hardware and software for the various internet purpose and coverage here, as it is a very vast and growing field.

Security: The security of the internet facilities is an important challenge in connecting a university to the internet. As have been repeatedly stated, the cost of the internet facilities and installation is enormous as such the issue of the security should be given utmost consideration.

- For the hardware security, they must be prevented from
 - theft and vandalization
 - environmental hazards such as thunder storms, rainstorms, etc.
- For the software security, they must be prevented from
 - unauthorized access and hacking
 - attack by virus, trojans, spywares and malicious malewares. For example, the presence of virus in a network can drags down the entire network and chokes up the bandwidth.



Fig 2 (colour online): Network Security (firewall)

Maintainability and Upgrade

A major problem in Nigeria today is lack of maintenance culture. Thus there are a number of cases where a project gradually comes to halt because the equipments could not be maintained or upgraded when necessary. The internet facilities require regular maintenance and even upgrading. For the maintenance, the staff responsible for the process need continuous monitoring and continuous action to keep the facilities working. In case there are parts to be replaced, the staff should act promptly and with standard materials. Similarly, whenever the need to upgrade arises which is very common because of the dynamic nature of ICT, this should be effected appropriately. However, owing to the present scenario where we import most of the internet facilities as well as the poor exchange rate of the Naira to the dollar, maintainability and upgrading of facilities pose a challenge to effectively connect our universities to the internet.

Prospects for Effective Connecting our Universities to the Internet.

The benefits of ICT in helping to improve the quality of life and the standard of living in the developed nations are enormous (Fansanya, 2000). Consequently, the United Nations (UN) in April 2000, initiated moves to assist Nigerian and other developing countries overcome obstacles that have kept them out of the IT revolution (Ogidan, 2000). On its part, one of the objectives of the Nigeria National Policy on S&T [Sec 3.16.2(b)] is to ensure that the country is part of the Global Information System with adequate communication and information facilities. This is in line with the objective of the National University Commission to make all Nigerian universities to be connected to the internet. This objective is being pursued by the Federal Government through empowering universities by donating some of these internet equipments through ETF and other ICT empowerment bodies. The success of this drive still depends on the aforementioned challenges. For example, a university management that do not understand the importance of ICT may end up poorly implementing the usage of the facilities from the federal government by giving out the personal computers/laptops for the project as gifts to individuals and thereby leaving the other facilities lie waste in some places in the university. The need to expose such managements to the relevance of ICT can then not be overemphasized.

In general, the challenges can be overcome by taking the following steps:

❑ Establishing Standard ICT Policy

The 21st university cannot function properly without an adequate provision of internet facilities. Apart from making available materials beyond a physical libraries and data beyond the physical laboratories, it can also be used for effective administration as an intranet (Akpojotor et. Al., 2010). For example, the academic planning unit and the Establishment division of the Delta State University can function more effectively with a good intranet service. Similarly, coordinating results between the University Senate, departments, and the Examinations and Records division can be enhanced by intranet service and this will have a far reaching benefits for the certificate and transcript procession. The possibility of an effective intranet service is only possible if there is a functional internet service. Therefore, the universities' management in developing countries must see the relevance of the internet not only as an information source but also as a means to provide effective administration and this should be their guiding philosophy when planning the ICT policy.

❑ Employing/Deploying of ICT experts in ICT

The relevance of the ICT to the university community which have been explained above makes it imperative that both in appointment of ICT unit Head and staff for the unit, only experts on ICT should be considered. Further, there should be an established and well funded means to develop the staff regularly through attending workshops, conferences, etc.

❑ Use of ICT personnel to execute ICT(i.e. Internet) projects

We have pointed out the remarkable difference in using external bodies to implement ICT policies and using the university staff by using the experience at the Delat State University as an example. This is not to say that all ICT jobs can be handled by the university ICT staff. The salient issue here is that in implementing the Ict policy, the university personnel should be used for those jobs they can handle.

❑ Funding by the private sector

ICT is very expensive as we have explained earlier and funding by government have not been adequate. There is need for universities in the developing countries to reach out to the private sector to support their ICT needs. This was recently epitomized when the Nigerian University Nsukka in partnership with pretty much every major IT company (Google Inc, MTN, Hewlett Packard Inc, Cisco Systems Inc, etc) operating in Nigeria to bring Internet connectivity of 1- 4 mbps per 1000 students and staff, and create 10-400 Terabytes of local data storage for students and faculty. This is currently the Africa's largest wireless network community

Conclusion

We have discussed the challenges that mar internet connection in Nigeria Universities both at general and local scales. We have also proffer some possible solutions. We hoped that the prospects outlined in this paper can help achieve a successful internet connection and be implemented at a level will meet the internet needs of these universities. Then we can reverse the old humble mockery of isolated researchers in S & T in developing countries, “Einstein can now be born in a jungle, ghetto, creek, etc” (Akpojotor et al., 2009).

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RADIOANALYSIS OF NATURAL KAOLIN USING NUCLEAR SPECTROSCOPY

C.E Mokobia

Department of Physics, Delta State University, Abraka

Email: mokobia_c@yahoo.com

Abstract

A radioanalysis of natural kaolin obtained from Ozanogogo Ika South was carried out using the Nigeria Research Reactor-1 (NIRR-1) with the aim of determining the radioelements present in the mineral and consequently the inherent natural radioactivity contained thereof. These parameters are needed for the estimation of radiological hazard indexes associated with the use of the material. The elemental characterization result showed the presence of aluminium, titanium and iron with concentrations $215300 \pm 1.2 \text{ mgkg}^{-1}$, $14200 \pm 8.0 \text{ mgkg}^{-1}$, and $1469 \pm 85 \text{ mgkg}^{-1}$ respectively as the major elements. Additionally, 17 other elements, in minor and trace quantities, including Thorium Th, a radioactive rare earth element and three others Lanthanum La, Dysprosium Dy and Lutetium Lu were identified. The presence of Thorium suggests the possibility of internal radioactivity in the rock, a necessary criterion for using a material for luminescence dating. Thus this work suggests the possibility of using kaolin for luminescence dating if properly developed for the purpose. The radionuclides detected in the gamma spectroscopic analysis belong to the Uranium-238 ^{238}U and Thorium-232 ^{232}Th series decay and the non series ^{40}K .

Key words: Radioanalysis, natural kaolin, radioelements, radionuclides luminescence dating.

Introduction

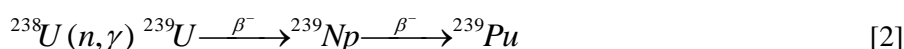
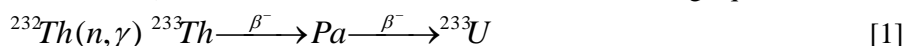
Kaolin (Chinese *kaoling*, high ridge”) or china clay is a pure, soft, white clay of variable but usually low plasticity that retains its white color when fired. The term kaolin may be extended to include other porcelain clays not discolored by firing. The material was first obtained from a hill called Kaoling and was sent to Europe in the early 18th century. Its chief constituent is the mineral kaolinite, a hydrous aluminum silicate, $\text{Al}_2\text{Si}_2\text{O}_5(\text{OH})_4$, formed by the decomposition of aluminum silicates, particularly feldspar (Truman and Thomas, 2008) the proportions of the compound components having been found to be 46% SiO_2 , 40% Al_2O_3 and 14% H_2O . Its structure may be flake-like, lathlike, fibre-like or hollow tube-shaped.

In the United States, the mineral is mined primarily in South Carolina, North Carolina, Georgia, Pennsylvania, and Alabama. Of all these, Georgia ranks first in production.

In Nigeria, it is found in Kebbi, Sokoto, Katsina, Kano, Jigawa, Plateau, Kogi and Kwara states in the North and Oyo, Ogun, Delta, Enugu and Abia states in the South. In Delta State, this mineral is found and mined predominantly in areas around Ozanogogo Ika South. It is also found in non commercial quantity around Ubulu-Ukwu, Aniocha South Local Government Area.

In its pure form, it is used in the manufacture of fine porcelain and china; impure varieties are used in making pottery, stoneware, and bricks (Truman and Thomas, 2008). The mineral is also used in the paper industry as fillers in the bulk. Its whiteness is employed in improving paper brightness and opacity. It is applied as white wares providing strength and plasticity in ceramics products. It is used to improve the optical, mechanical and rheological properties of paints and it is also used in pharmaceuticals for the production of human and veterinary medicinals for the treatment of digestion problems. These varying applications highlight the importance of this mineral in a developing nation like Nigeria.

Of recent, the radioactive contamination of geological materials has attracted great attention especially as these naturally occurring radioactive materials (NORM) are known not only to have reached hazardous levels (El – Dine et al, 2001). In fact they have been found to be capable of resulting in much larger radiological exposure to the public relative to that caused by the nuclear industry for instance (Mokobia et al, 2006). Discussions of the health implications of this kind of radiation on the populace must be translated to relevant control measures. This is one of the main focuses of the Nigerian Nuclear Regulatory Agency (NNRA). One aspect of this health implication has to do with ^{222}Rn (radon) the only gaseous member of the $^{238}\text{U}/^{226}\text{Ra}$ (Uranium) decay series part of which is shown in Figure 1 (Mokobia, 2004a; 2008a). This short-lived radioactive gas (0.83days) decays into much shorter-lived ^{218}Po (Polonium) and ^{214}Pb (Lead) emitting α particle which causes cancer of the lungs if breathed in (Mokobia, 2004b). The other aspect has to do with the accumulation of ^{239}Pu (Plutonium) when ^{238}U interacts with atmospheric neutrons through (n, γ) nuclear reactions. These reactions result in the production of fissile materials (Mokobia et al, 2006) ^{233}U and ^{239}Pu as illustrated in the following equations:



This gradual production of ^{239}Pu presents a crucial but unrecognized problem in radiation protection especially as it is reportedly toxic even in minute quantities. The neutrons in such nuclear reactions are primarily produced by the bombardment of oxygen/ nitrogen nuclei in the atmosphere by cosmic rays. Such nuclear reactions may be represented by the nuclear equation:



Furthermore, several other radionuclides in the decay chain are known to be radiotoxic (Ahmed, 2004). Their presence in water for instance can constitute a serious health risk (Rowland, 1993). Exposure to very high levels of radium for a long period of time may result in cancers of the bones and of the nasal cavity (Wren et al, 1985).

In this study, a radioanalysis of natural kaolin obtained from Ozanogogo Ika South was carried out using a gamma spectrometer and the Nigeria Nuclear Research Reactor.

The purpose was to determine the radioelements present in the mineral and consequently the inherent natural radioactivity contained thereof. These are vital parameters needed for the estimation of radiological hazard indexes associated with the use of this highly valuable material consequent upon the inherent natural radioactivity.

Materials and Methods

Samples of the mineral under study were collected from five locations around Ozanogogo (latitude $6^{\circ} 8' - 6^{\circ} 10' \text{N}$ and longitude $6^{\circ} 6' - 6^{\circ} 17' \text{E}$) in Ika South Local Government area of Delta State (Olobaniyi et al, 2007). On collection, these samples were securely tied in dark cellophane bags and properly labeled. The as-collected samples were washed, cleaned with acetone and air dried to constant weight at room temperature in the sample preparation laboratory at the Centre for Energy Research and Development (CERD) Obafemi Awolowo University Ile-Ife. They were then separately crushed using a thoroughly cleaned crusher and separated into two portions each of which was separately stored in a securely tied dark cellophane bag.

For the purpose of the gamma spectroscopic investigation, one of the stored portions of the samples was used. Known masses of the sample were weighed into Marinelli beakers and firmly sealed. The sealed containers were left for 28 days so that secular equilibrium between the parent and daughter nuclides present in the samples hitherto disturbed during the sample collection was restored in line with conventional practice (IAEA, 1989; Olomo, 1990; Mokobia et al., 2003 & 2006) preparatory for gamma counting.

Gamma counting was performed using a highly efficient NaI (TL) Scintillation detector located at CERD, Obafemi Awolowo University Ile-Ife. Energy and efficiency calibrations were carried out using mixed gamma-ray standard sources, which were obtained from the National Bureau of Standards of the United States. Each sample as well as the mixed standard source and the background were counted for 10 hrs. It

was ensured that the sample containers and that of the background and standard are of the same configuration so as to minimize error. Spectra evaluation was carried out using a PC based SAMPO 90 computer program capable of matching the γ -energies at various levels to a library of possible isotopes.

The specific activity of each identified radionuclide in the samples was calculated comparatively using the relation (Mokobia, 2008b):

$$A_s = \frac{N(E_\gamma)_s M_d A_d}{N(E_\gamma)_d M_s} \quad [4]$$

A_s being the specific activity in Bqkg^{-1} of the identified nuclide having a net photopeak area $N(E_\gamma)_s$ contained in a sample of mass M_s in kg, A_d the activity of this nuclide as contained in the mixed standard of mass M_d in kg and $N(E_\gamma)_d$ the net photopeak area of the said nuclide. The uncertainty in each value of specific activity to be obtained for each of samples was determined using the equation:

$$\Delta A_s = A_s \left\{ \left| \frac{\Delta N(E_\gamma)_s}{N(E_\gamma)_s} \right|^2 + \left| \frac{\Delta M_d}{M_d} \right|^2 + \left| \frac{\Delta A_d}{A_d} \right|^2 + \left| \frac{\Delta N(E_\gamma)_d}{N(E_\gamma)_d} \right|^2 + \left| \frac{\Delta M_s}{M_s} \right|^2 \right\}^{0.5} \quad [5]$$

ΔA_s , ΔM_d , ΔA_d , $\Delta N(E_\gamma)_d$ and ΔM_s represent the uncertainties in these quantities already defined.

For the purpose of carrying out the radioanalysis, the remaining portion of the stored sample was used. 150 mg each of the samples were separately weighed into heat sealed, labeled small plastic sachets in the sample preparation laboratory of the Nigeria Research Reactor-1 (NIRR-1) sited at the Centre for Energy Research and Training (CERT), Ahmadu Bello University, Zaria. The sealed sachets were thereafter encapsulated into irradiation vials preparatory to irradiation using thermal neutrons in the reactor.

Two irradiation regimes (A1) and (A2) and two counting strategies (M1 and M2) after each activation were adopted. In the first irradiation protocol, the samples were separately sent into the reactor through a multifunctional transfer system (the pneumatic transfer system) connected to three small inner irradiation channels and a small outer irradiation channel (Figure 2). Irradiation was done at a thermal neutron flux of $2 \times 10^{11} \text{ ncm}^{-2}\text{s}^{-1}$ for a period of 120 s. Thereafter the irradiated samples were returned to the ejector via the same route. The irradiated samples were then counted using a well calibrated HPGe gamma-ray spectrometer after a cooling time of 120 s at detector to source distance of 15 cm in order to eliminate errors due to coincident losses. Recounting was done at a shorter source to detector distance of 1.8 cm 2 h later so as to be able to detect medium-lived radioelements such as ^{24}Na (14.96 h), ^{56}Mg (2.56 h) etc.

For the second irradiation protocol, the samples were irradiated for 6 h. This was preceded by counting after a cooling time of 270 h for 1800 s. Another series of counting followed after a cooling period of 898 h for 5000 s. This last counting was aimed at reducing interference and scattering from lower and medium half-lived radioisotopes.

In the quantification of the radioelements detected in the samples in either of the irradiation regimes and spectrometric measurements, the concentration of the radioisotope i, produced after irradiating the sample for a time t was determined using the multi-purpose gamma-ray analysis software WinSPAN-2004 (WangLiu, 2004). The basic equation for determining the detector response following the irradiation of a

sample in a thermal neutron flux ϕ as given in Balogun et al. (1996) is:

$$D = P_\gamma a_f m \epsilon \sigma \phi \frac{N_A}{A_i} \left(\frac{1 - e^{-\lambda t}}{\lambda} \right) e^{-\lambda t_w} (1 - e^{-\lambda t_c}) \quad [6]$$

D is the detector response (counts/ sec), ϵ is the detection efficiency at the gamma –ray energy of interest, P_γ is the gamma – ray yield of energy of interest, m is the mass of the element of interest (kg), a_f is the weight fraction (abundance) of the isotope of atomic mass A_i , λ is the decay constant of the

produced radioisotope, σ_i is the microscopic neutron capture cross section (barn) for the reaction that produces the radioisotope, ϕ is the neutron flux (neutron $\text{m}^{-2}.\text{s}^{-1}$), N_A is the Avogadro's number, t is the irradiation time, t_w is the time between end of irradiation and the start of counting, t_c is the counting time. The first parameter in parenthesis on the right hand side of this equation is a correction term accounting for decay during irradiation, the exponential following is the correction for the waiting time before counting started while the last term in parenthesis takes care of the correction for the counting since the element also decays even while counting.

Results and Discussions

The radionuclides detected in the gamma spectroscopic analysis belong to the ^{238}U and ^{232}Th series decay and the non series ^{40}K . As shown in Table 1, the ranges of the specific activities of these primordial radionuclides are $(285.96 \pm 6.97 - 288.80 \pm 3.43) \text{ Bqkg}^{-1}$ $(299.11 \pm 3.89 - 299.82 \pm 6.34) \text{ Bqkg}^{-1}$ and $(268.63 \pm 2.50 - 271.82 \pm 3.64) \text{ Bqkg}^{-1}$ respectively. The contributions of these nuclides in the overall radioactivity Contained in this geological sample are 33%, 35% and 32% respectively.

The elemental characterization (Table 2) showed the presence of 20 radioelements. These include the major elements aluminium, titanium and iron with concentrations $215300 \pm 1.2 \text{ mgkg}^{-1}$, $14200 \pm 8.0 \text{ mgkg}^{-1}$, and $1469 \pm 85 \text{ mgkg}^{-1}$ respectively. Additionally, 17 other elements, in minor and trace quantities, including Th, a radioactive rare earth element and three others La, Dy and Lu were identified. The presence of Th suggests the possibility of internal radioactivity in the rock. The possession of inherent internal radioactivity by a geological sample is indicative of the applicability of the material in luminescence dating. Thus this work suggests the possibility of using kaolin for luminescence dating if properly developed for the purpose.

Conclusion

The elemental characterization of natural kaolin using neutron activation analysis showed the presence of aluminum, titanium and iron with concentrations $215300 \pm 1.2 \text{ mgkg}^{-1}$, $14200 \pm 8.0 \text{ mgkg}^{-1}$, and $1469 \pm 85 \text{ mgkg}^{-1}$ respectively as the major elements. Additional 17 other elements, in minor and trace quantities, including Th, a radioactive rare earth element and three others La, Dy and Lu were identified. The presence of Th suggests the possibility of internal radioactivity in the rock, a necessary criterion for using a material for luminescence dating. Thus this work suggests the possibility of using kaolin for luminescence dating if properly developed for the purpose. The radionuclides detected in the gamma spectroscopic analysis belong to the ^{238}U and ^{232}Th series decay and the non series ^{40}K .

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Table 1: Specific activities of nuclides detected in natural kaolin

Sample Identity	^{40}K (Bq kg ⁻¹)	Variation	$^{236}\text{U}/^{226}\text{Ra}$ (Bq kg ⁻¹)	Variation	$^{232}\text{Th}/^{228}\text{Ra}$ (Bq kg ⁻¹)	Variation
K1	270.11 ± 4.00	0.158	287.71 ± 4.00	0.072	299.80 ± 5.52	-0.298
K2	268.63 ± 2.50	1.638	288.80 ± 3.43	-1.018	299.48 ± 6.45	0.042
K3	271.82 ± 3.64	-1.552	287.73 ± 3.43	0.052	299.32 ± 3.89	0.182
K4	270.46 ± 6.42	-0.192	285.96 ± 6.97	1.822	299.11 ± 3.89	0.392
K5	270.32 ± 3.59	-0.052	288.71 ± 7.16	-0.928	299.82 ± 6.34	-0.318
Mean	270.27 ± 20.15		287.78 ± 24.99		299.50 ± 25.99	

Table 2: Elemental concentrations of radio elements in natural kaolin ($\text{mgkg}^{-1} \pm \%$) NAA

Element	Concentration	LOD	Element	Concentration	LOD
AL	215300 ± 1.2	17	Sc	23.9 ± 0.2	0.2
Ti	14200 ± 5.3	5200	Cr	164 ± 7	23
V	219 ± 8.2	15	Fe	1469 ± 85	829
Na	216.5065 ± 16.0	15	Co	4.1 ± 1.4	3.0
Mn	22.3692 ± 12.3	0.9	Zn	BDL	3.0
Eu	5.1287 ± 5.0		Ba	264 ± 85	264
Dy	11.1658 ± 5.9	0.7	Ce	BDL	14
La	126 ± 0.9	0.2	Lu	0.55 ± 0.04	0.1
Sm	BDL	0.1	Ta	3.8 ± 0.5	1.0
Br	1.8 ± 0.4	3.0	Hf	8.6 ± 0.6	1.1
Yb	5.9 ± 0.5	0.9	Th	28.8 ± 0.8	1.2

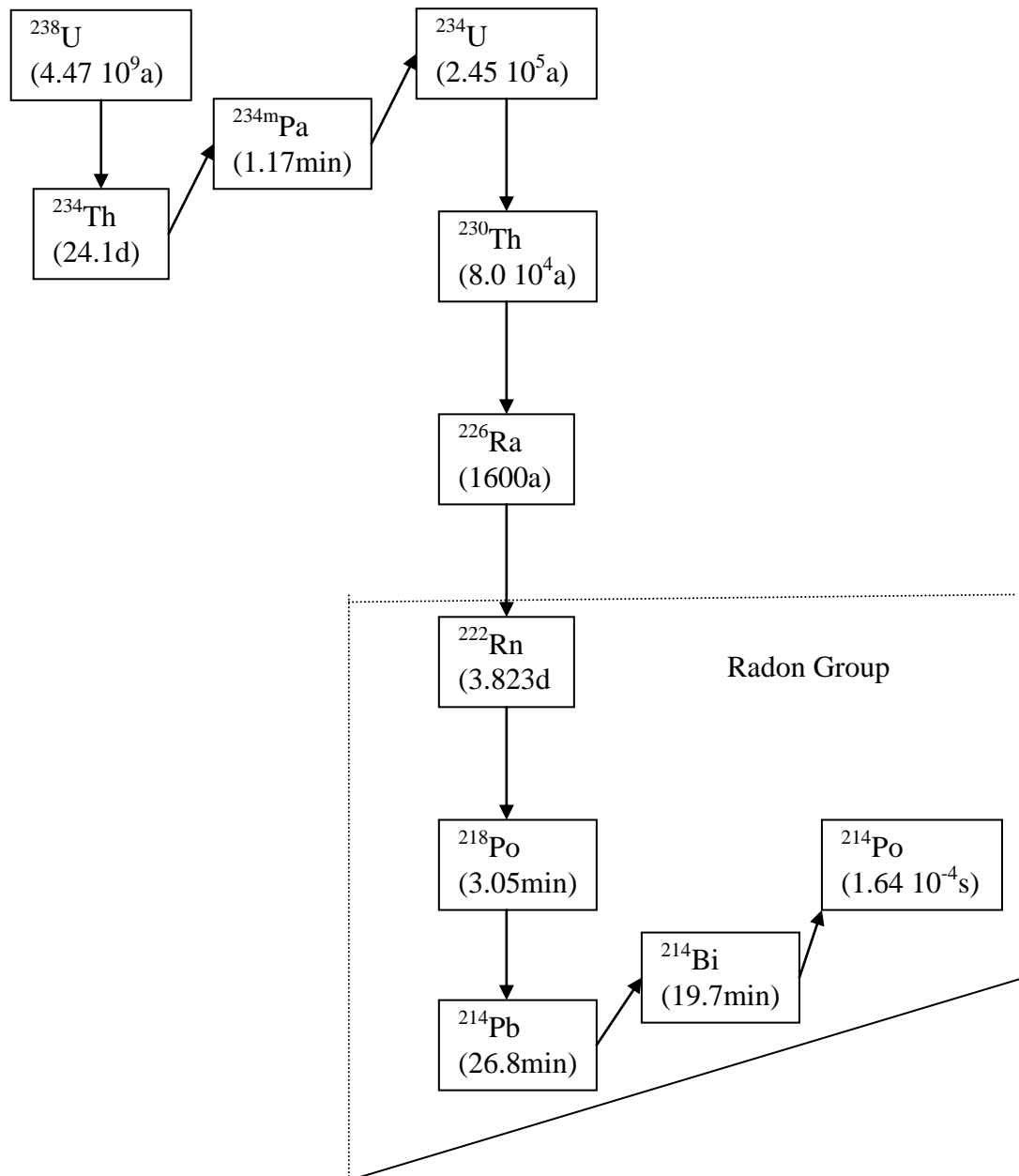
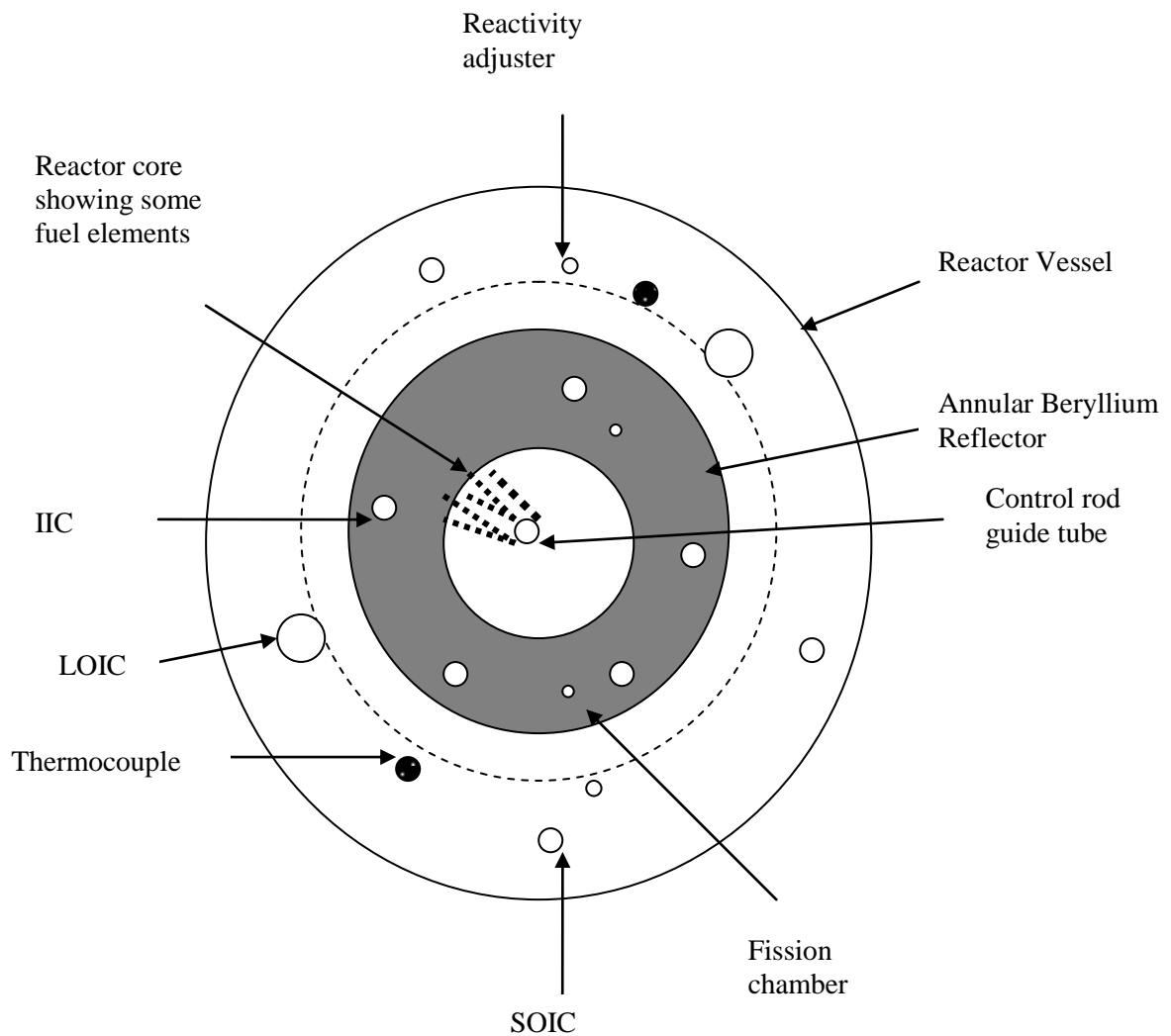


Fig. 1: ^{238}U decay series showing ^{222}Rn and its progenies (after Mokobia, 2004a; 2008)



OIC – Outer irradiation channel, IIC – Inner irradiation channel

Fig. 2.: Core configuration of the NIRR-1 showing the irradiation channels (after Jonah et al., 2005)

BIOSORPTION OF SELECTED DIVALENT METAL IONS FROM AQUEOUS SOLUTIONS BY YAM (*Dioscorea Rotundata Poir*) TUBER PEEL WASTE

P.M. Eguvbe, C.M.A. Iwegbue, G.E. Nwajei and S.H.O. Egbob

Department of Chemistry Delta State University, Abraka, Nigeria.
(Phone: +2348067562013; e-mail: eguvbepeter@yahoo.com)

Abstract

The biosorption of five divalent metal ions, Co(II), Cu(II), Ni(II), Cd(II) and Pb(II), onto ground yam (*Dioscorea rotundata poir*) tuber peel waste biomass over a wide range of reaction conditions and equilibrium-sorption kinetics was studied. The batch experiments show that pH 4-5 was the best range for the sorption of the metal ions onto the biomass. Time-dependent experiments for the metal ions showed that for the five metals examined, binding to the yam tuber peel waste biomass was rapid and occurred within 25min and completed within 1hr. The sorption process was examined by means of *Langmuir* and *Freundlich* isotherms. According to the evaluation using *Langmuir* equation, the monolayer sorption capacity obtained was 2.27mg/g Co(II), 1.19mg/g Pb(II), 0.89mg/g Cu(II), 0.86mg/g Ni(II) and 0.85mg/g Cd(II). The kinetics of the sorption mechanism was evaluated using the pseudo-first order rate model and the pseudo-second order rate model. The results revealed that pseudo-second order model provides a more appropriate description of the metal ion sorption process of Co(II), Cu(II), Ni(II), Cd(II) and Pb(II) onto yam tuber peel waste biomass. The results from these studies indicated that yam tuber peel waste biomass could be employed in the removal of toxic and valuable metals from industrial effluents.

Keywords: Biosorption, heavy metals, industrial effluents, yam tuber peel waste.

Introduction

Rapid industrialization has led to discharge of effluent containing heavy metals into the atmosphere, land and water bodies. The presence of heavy metals in the environment has become a major threat to plant, animal and human life due to their bioaccumulating tendency and toxicity and therefore must be removed from municipal and industrial effluents before discharge [1].

Physico-chemical methods such as chemical precipitation, lime coagulation, ion exchange, solvent extraction, reverse osmosis, chemical oxidation or reduction, electrochemical treatment, evaporative recovery, filtration and membrane technologies have been widely used to remove heavy metal ions from industrial waste water [2-6]. The processes may be ineffective or expensive and non environment friendly, especially when the heavy metal ions are in solutions containing in the order of 1-100mg of dissolved heavy metal ions/L [7]. Moreover, the disadvantages like incomplete metal removal, high reagent and energy requirements, generation of toxic sludge or other waste products that require careful disposal has made it imperative for a cost effective treatment method that is capable of removing heavy metals from aqueous effluents.

Biological methods such as biosorption/bioaccumulation for the removal of heavy metal ions may provide an attractive alternative to physico-chemical methods, as they are more environment friendly and economically feasible [8].

Biosorption of heavy metals by microbial biomass has been recognized as a potential alternative to the existing physico-chemical technologies for detoxification and recovery of toxic and valuable metals from waste water [7],[9]. Many micro-organisms; fungi, yeast, bacteria and algae have been identified as superior candidates for bioremediation owing to their ability to sequester cationic and anionic metallic species [10-12]. The major advantages of biosorption over conventional treatment methods include the treatment of large volume of effluents with low concentration of pollutants, low cost, high efficiency, minimization of chemical and/or biological sludge, no additional nutrient requirement, regeneration of biosorbent, and possibility of metal recovery [13],[9],[4].

Recent biosorption experiments have focused attention on waste materials, which are by products or the waste materials from large-scale industrial processes. For example waste mycelia available from fermentation processes, olive mill solid residue [14], activated sludge from sewage treatment plants, biosolid [15]. In recent times a great deal of interest has been given to the utilization of agricultural by-products as adsorbents for the removal of trace amounts of toxic and valuable heavy metals from municipal and industrial wastewater effluents, particularly because of the low cost, high availability of these materials, no need for complicated regeneration process and they are capable of binding to heavy metals by adsorption, chelation and ion exchange [16-19].

Research in the use of agricultural by-products has included metal binding studies with, apple waste, wheat and rice, oat fibre, sugarcane bagasse, sawdust, peanut skins, sunflower, peat moss, *medicago sativa* (Alfalfa), walnut waste and cassava waste etc, [20-2] and [17-19].

The objective of the present study is to determine the suitability of the use of *Dioscorea rotundata* *poir* (Yam) tuber peel wastes as a biosorbent for valuable and toxic metals from aqueous media and also to assess the effect of reaction conditions on the adsorption isotherm and kinetic processes during the sorption of metal ions.

Experimental Part

Materials: Forty (40) tubers of white yam (*Dioscorea* Series) were collected from Emevor market in Delta State Nigeria. The tubers were washed with distilled deionized water and allowed to air dry. The dried tubers were cut into 6cm and carefully peeled with a kitchen knife to obtain the yam peel wastes. The peelings were sun dried for five days. The dried samples were ground using a mechanical grinder and sieved through a standard screen to obtain a particle size of 90 μ m and stored in a plastic container for analysis. The purification and activation was carried out according to the previous work of [1]. 500g of finely divided biomass was activated and at the same time purified by soaking in excess 0.3M HNO₃ for 24h to remove any metals and biomolecules, followed by washing thoroughly with deionized H₂O until a pH of 7.1 \pm 0.1 was attained and then air-dried. The air-dried activated biomass was then washed with deionized water and re-suspended in 1.0M hydroxylamine to remove all o-acetyl groups. To remove all other soluble materials, the biomass was washed with deionized water and centrifuged at 3000rpm for five minutes using a portable refrigerated test tube centrifuge. The supernatants obtained were discarded and the purified biomass cake obtained was dried at room temperature.

Effect of Metal-Ion Concentration: Several standard solutions with concentrations of 10,20,30,40,50,60,70,80,90 and 100mg/l were made from spectroscopic grade standards of Cd(II) (from Cd(NO₃)₂ · 4H₂O), Ni(II) (from Ni(NO₃)₂), Pb(II) (from Pb(NO₃)₂), Cu(II) (from CuSO₄), and Co(II) (from Co(NO₃)₂ · 6H₂O). The metal solutions made separately were adjusted to pH 5.0 with conc. HCl. 50ml of each metal ion solution was added to accurately weighed 5g activated/purified biomass in different flasks and agitated gently for 2hrs to ensure that equilibrium was achieved. At the end of the time, the suspension was filtered through whatman No. 45 filter paper and centrifuged at 3000rpm for 5min. The supernatants were analyzed for heavy metals.

Effect of Contact Time: This was assessed by the addition of metal solutions (Cd(II), Ni(II), Pb(II), Cu(II) and Co(II)) (2.50mg in 50ml water) adjusted to pH 5 into 5g activated/purified biomass and was shaken for time intervals of 5,10,15,20,25,30,40,50 and 60min. At the end of time interval, the suspension was filtered and centrifuged at 3000rpm for 5min and the metal content was analyzed as described above [22].

Effect of pH on sorption: This was assessed by adjusting the pH of the metal solutions Cd(II), Ni(II), Pb(II), Cu(II) and Co(II) (2.50mg in 50ml water) to pH 2.0, 3.0, 4.0, 5.0, 6.0, 7.0, 8.0 and 9.0 by addition of either concentrated HCl or NaOH and then adding it into 5g activated/purified biomass and shaken for 1h. The suspensions were filtered through whatman 45 and then centrifuged at 3000rpm for 5min. The metal content was analyzed using Flame Atomic Absorption Spectrophotometer.

Data Evaluation: The amount of Cd(II), Ni(II), Pb(II), Cu(II) and Co(II) ions removed by the biomass during the series of batch investigations were determined using a mass balance equation (Equ.1).

$$q_e = \frac{v}{m}(C_0 - C_e) \quad (1)$$

where q_e = metal ion concentration on the biomass (mg/g) at equilibrium; C_e = metal ion concentration in solution (mg/L) at equilibrium; C_0 = initial metal ion concentration in solution (mg/L); v = volume of initial metal ion solution used (L); m = mass of biomass used (g).

Equilibrium Isotherms: Two adsorption models were used to fit the experimental data: the Langmuir and the Freundlich models. The *Langmuir* (L) equation was chosen for the estimation of maximum adsorption capacity, corresponding to complete monolayer coverage on the biomass surface, as expressed by Equ. 2.

$$q_e = \frac{q_m K_L C_e}{1 + K_L C_e} \quad (2)$$

Where K_L = *Langmuir* isotherm constant (L/g) and q_m = *Langmuir* monolayer adsorption capacity (mg/g). The linearized form of the above equation is given in Equ.3.

$$\frac{C_e}{q_e} = \frac{1}{q_m K_L} + \frac{C_e}{q_m} \quad (3)$$

The experimental data were fitted into equation (3) by plotting C_e/q_e against C_e .

The favourability of adsorption of the five metal ion on the biomass was tested using the essential feature of the *Langmuir* isotherm model expressed in terms of a dimensionless constant called separation factor (S.F) proposed by [23]. The separation factor S.F, is defined by the following relationship.

$$S.F. = \frac{1}{1 + K_L C_0} \quad (4)$$

where; K_L = *Langmuir* isotherm constant; C_0 = initial metal ion concentration. The parameter indicates the shape of the isotherm as follows: $SF > 1$ unfavourable isotherm; $SF = 1$ linear isotherm; $SF = 0$ irreversible isotherm; $0 < SF < 1$ favourable isotherm.

The *Freundlich* model was used to estimate the adsorption intensity of the sorbent toward the biomass and is given in Equ.5.

$$q_e = K_F (C_e)^{1/n} \quad (5)$$

Where; q_e = the adsorption density (mg of metal ion adsorbed/g biomass); C_e = the concentration of metal ion solution at equilibrium (mg/L); K_F and n are the *Freundlich* constants. The value of n indicates the affinity of the sorbent towards the biomass. Equation 5 is conveniently linearized by taking the natural logarithm of both sides as:

$$\ln q_e = \ln K_F + 1/n \ln C_e \quad (6)$$

A plot of $\ln q_e$ against $\ln C_e$ that yields a straight line indicates the confirmation of the *Freundlich* adsorption isotherm. The constants $1/n$ and $\ln K_F$ can be determined from the slope and intercept respectively.

Kinetic treatment of experimental data was investigated by applying the pseudo-first order expression given by Langergren [24] and pseudo-second order [25]. The linear form of Langergren's pseudo-first order model is given by the equation.

$$\ln (q_e - q_t) = \ln q_e - K_t t \quad (7)$$

where q_e = mass of metal adsorbed at equilibrium (mg/g), q_t = mass of metal adsorbed at t (mg/g), K_t = equilibrium rate constant. A linear plot of $\ln (q_e - q_t)$ versus t confirms the model.

The pseudo-second order equation described by [25] for the sorption system of divalent metal ions using sphagnum moss plant was adopted. The linear form of Ho's pseudo-second order model is generally expressed as follows:

$$t/q_t = 1/h_o + 1/q_e t \quad (8)$$

where, q_t = the amount of divalent metal ion on the biomass surface (mg/g) at any time (t); q_e = the amount of divalent metal ion sorbed at equilibrium (mg/g), h_o = the initial sorption capacity (mg/g min). The initial sorption rate, h_o , is defined as Equ.9.

$$h_o = K_2 q_e^2 \quad (9)$$

where; K_2 is the pseudo-second order rate constant (g/mg min). A linear plot of t/q_t against t confirms the model.

Results and Discussion

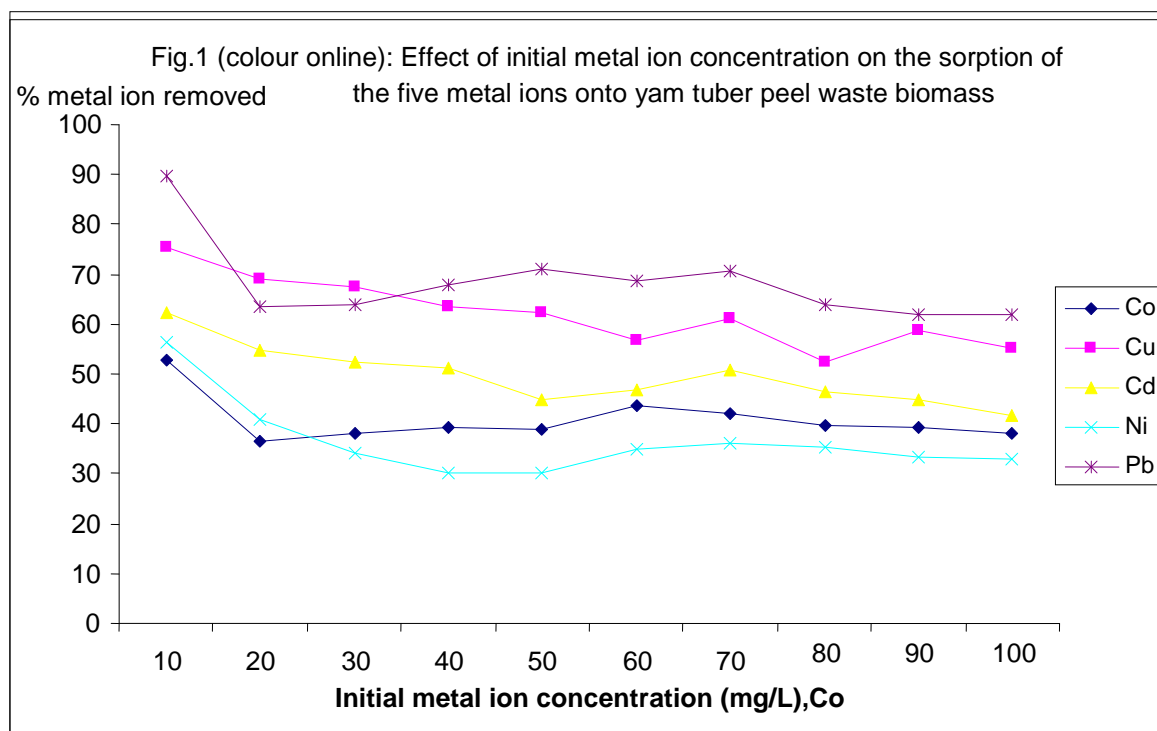
Effect of Metal-Ion Concentration

The results of the uptake of Co(II), Cu(II), Cd(II), Ni(II) and Pb(II) ions onto yam tuber peel waste biomass are at various initial metal ion concentration presented in Table 1. The sorption capacity increased with increasing concentration of metal ion from 10 to 100mg/L and a biomass dose of 5.0g. The sorption capacity of the metals followed the order Pb(II)>Cu(II)>Cd(II)>Co(II)>Ni(II).

Table 1: Effects of Concentration on Metal Ion Removal

Initial metal ion Concentration Co (mg/L)	Amount of metal adsorbed (mg/g), q_e				
	Co	Cu	Cd	Ni	Pb
10	0.053	0.075	0.062	0.056	0.090
20	0.073	0.138	0.110	0.082	0.127
30	0.114	0.203	0.157	0.102	0.191
40	0.156	0.253	0.205	0.121	0.271
50	0.194	0.311	0.224	0.152	0.356
60	0.261	0.341	0.281	0.210	0.413
70	0.294	0.428	0.355	0.252	0.495
80	0.316	0.418	0.372	0.284	0.509
90	0.352	0.530	0.403	0.301	0.559
100	0.382	0.552	0.416	0.333	0.621

However, the actual percent removal of the metal ions from solution was found to decrease with increase in initial metal ion concentration (fig. 1). This may be due to the fact that at lower concentrations almost all the ions were adsorbed very quickly, and further increase in initial metal ion concentrations led to saturation of the biomass surface. Similar type of adsorption have been reported [26],[1],[19] and [22].

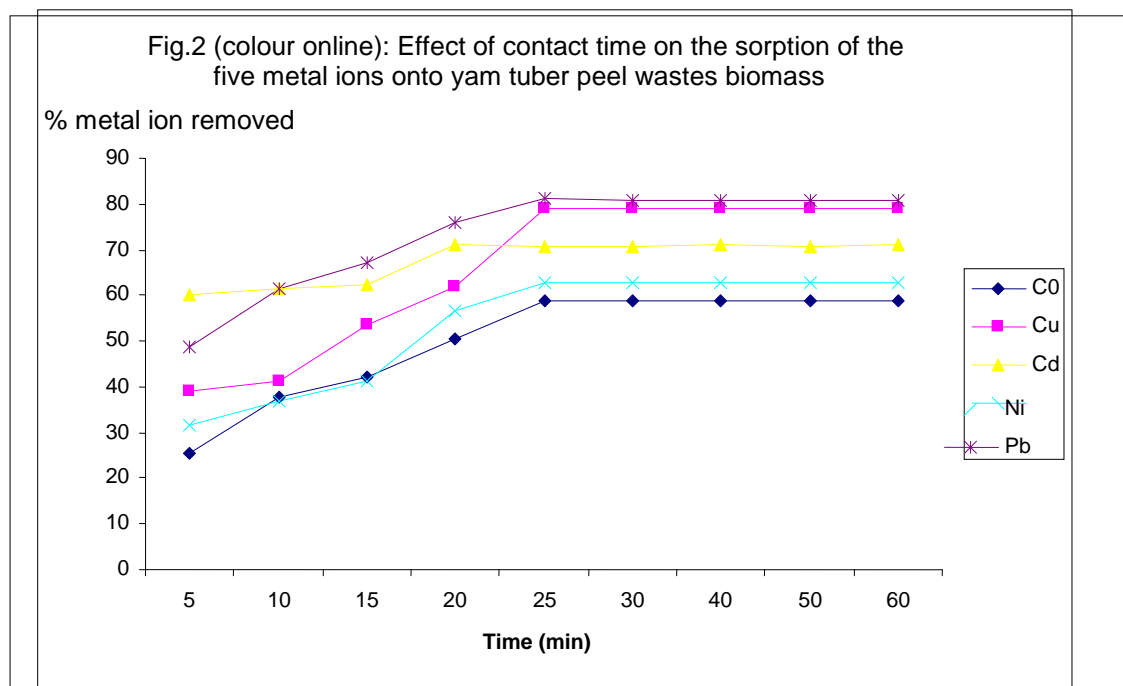


Effect of Contact Time

The data for time-dependency for the adsorption of metal ions onto yam tuber peel waste is presented in Table 2 and Fig.2. As the contact time was increased from 5 to 20 min, the amount of metal ions removed by the biomass increased, until a contact time of 25min was reached, thereafter, the amount of metal ion removed remained fairly constant. Within the first 5-25min, the biomass was capable of removing over 55-80% of each metal ion. Optimum adsorption of all the metals were achieved within 20-25min. Since all soluble components were removed during washings, the adsorption could only have taken place by the yam tuber peel waste biomass. The rapid adsorption of metal ions to the biomass indicated that adsorption might have been taking place on the cell wall of the biomass. The relative fast removal also indicates that physisorption as well as chemisorption processes are involved in the reaction between metal ions and yam tuber peel waste biomass. Similar adsorption trends have been reported for other biomasses [19] and [22].

Table 2: Effects of Contact Time on Metal Ion Removal

Contact Time (min.)	Co	Cu	q_e (mg/g) Cd	Ni	Pb
5	0.025	0.039	0.060	0.032	0.049
10	0.038	0.041	0.061	0.037	0.061
15	0.058	0.054	0.062	0.041	0.067
20	0.051	0.062	0.071	0.057	0.076
25	0.059	0.079	0.071	0.063	0.081
30	0.059	0.079	0.071	0.063	0.081
40	0.059	0.079	0.071	0.063	0.081
50	0.059	0.079	0.071	0.063	0.081
60	0.059	0.079	0.071	0.063	0.081

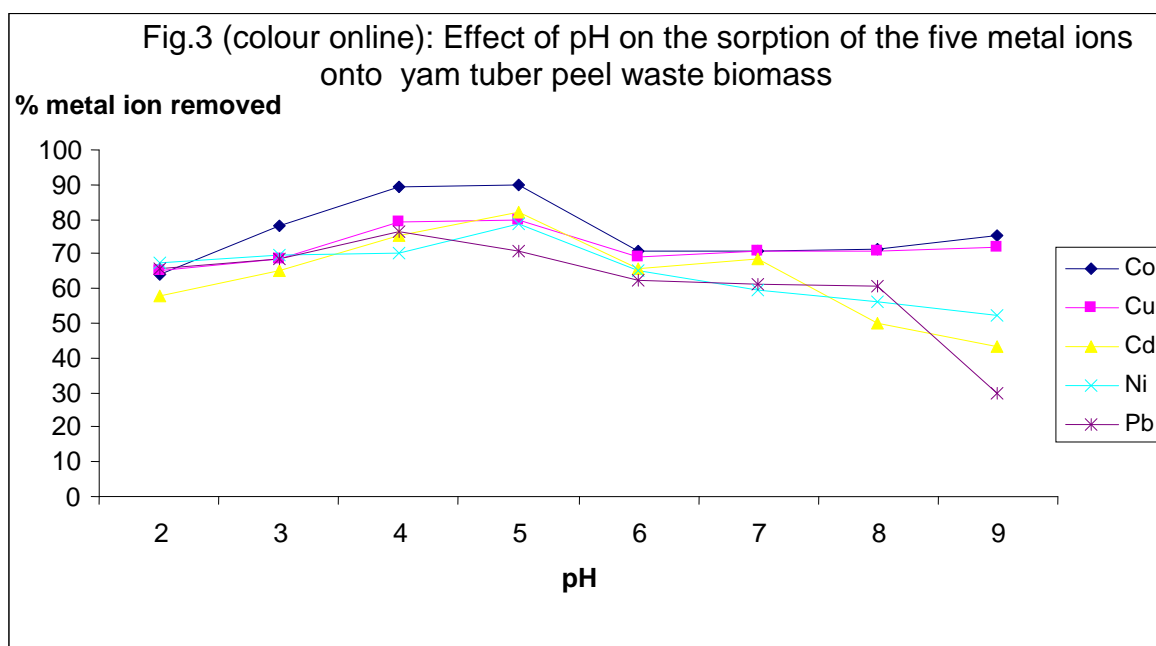


Effect of pH

The pH-dependence data for the sorption of the five metals under investigation are presented in Table 3 and Fig.3. The data showed that Co(II), Cd(II), Ni(II), Cu(II) and Pb(II) sorption increased as pH increased from 2 to 5, with optimum adsorption of 75-90% occurring between pH 4 and 5. Above pH 5, a gradual decrease in the amount of metal ions removed by the biomass was observed. In general, pH-dependency study showed that the sorption for the five metal ions onto yam tuber peel waste biomass were similar, increasing as pH increased and decreasing as pH approached 9.

Table 3: Effects of pH on Metal Ion Removal

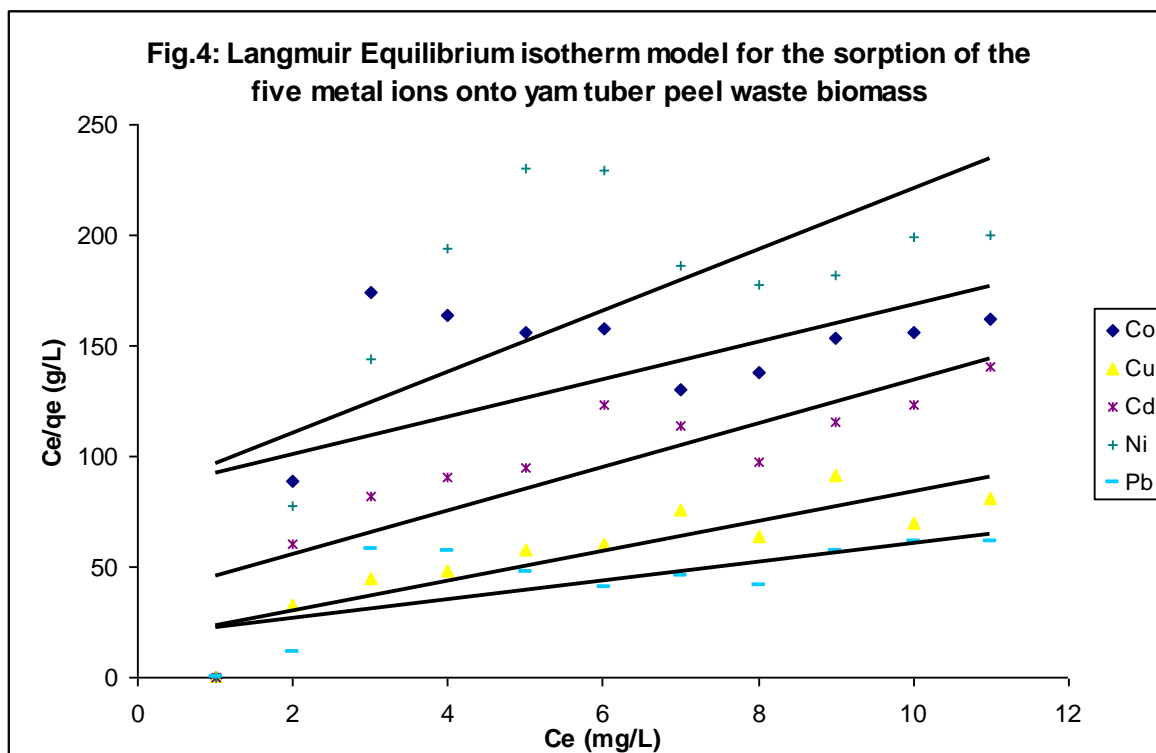
pH	q_e (mg/g)				
	Co	Cu	Cd	Ni	Pb
2	0.064	0.065	0.058	0.067	0.066
3	0.078	0.069	0.065	0.070	0.068
4	0.089	0.079	0.075	0.070	0.076
5	0.090	0.080	0.081	0.079	0.071
6	0.071	0.069	0.066	0.065	0.062
7	0.071	0.071	0.068	0.059	0.061
8	0.071	0.071	0.050	0.056	0.061
9	0.076	0.072	0.043	0.052	0.030



This effect suggested that the adsorption mechanism for the metals investigated might be an ion-exchange type process. Again, the adsorption mechanism for these metals were stable and rapid, which implies that adsorption is taking place on the cell wall surface of the yam tuber peel waste biomass. This trend in pH-dependency suggests that by reducing the pH, the bound metal ions could be desorbed and the spent biomass regenerated [16-18] and [27-28]. Once the metals are recovered, the biodegradable biomass can be used as soil amendment materials in agricultural lands.

Langmuir Isotherm

A plot of specific sorption (C_e/q_e) against the equilibrium concentration (C_e) is presented in Fig.4, and the linear isotherm parameters, q_m , K_L and the coefficient of determination (R^2) are presented in Table 4.



The R^2 values suggested that the *Langmuir* isotherm provides a good model of the sorption system. The sorption capacity, q_m , which is a measure of the maximum adsorption capacity corresponding to complete monolayer coverage showed that the yam tuber peel waste had a higher mass capacity that follows the order, $\text{Co(II)} > \text{Pb(II)} > \text{Cu(II)} > \text{Ni(II)} > \text{Cd(II)}$. The adsorption coefficient, K_L , which is related to the apparent energy of sorption follows the order, $\text{Cu(II)} > \text{Pb(II)} > \text{Cd(II)} > \text{Ni(II)} > \text{Co(II)}$. This showed that the energy of adsorption is not very favourable to Cu(II) probably due to its large ionic radius, hence not all binding sites may be available to Cu(II). Similar order has been by [1] for different metal ion sorption on fluted pumpkin waste biomass. The separation factor, SF, for the five divalent metal ion are less than 1 indicating that yam tuber peel waste biomass is an excellent adsorbent for the five metal ions. The SF values follow the order: $\text{Co(II)} > \text{Ni(II)} > \text{Cd(II)} > \text{Pb(II)} > \text{Cu(II)}$, indicating that in a mixed metal ion system, Pb(II), Cd(II), Cu(II) and Ni(II) will compete for binding sites faster than Co(II). This observed separation factor indicates that high concentration of Pb(II), Cd(II), Cu(II), Co(II) and Ni(II) ions in an effluent will not be a limiting factor in the ability of yam tuber peel waste to sorb these metal ions. Similar separation factor has been reported [1].

Table 4: Linear Langmuir Isotherm Parameters

Metal ions	q_m (mg/g)	K_L (L/g)	R^2	SF
Co(II)	2.27	0.003	0.119	0.971
Cu(II)	0.89	0.030	0.849	0.769
Cd(II)	0.85	0.020	0.818	0.833
Ni(II)	0.86	0.008	0.311	0.926
Pb(II)	1.19	0.025	0.403	0.800

Freundlich Isotherm

The linear *Freundlich* isotherms for the sorption of the five divalent metals onto yam tuber peel waste biomass is presented in Fig.5, and Table 5 shows the linear *Freundlich* sorption isotherm constants and the coefficients of determination (R^2). Based on the R^2 values, the linear form of the *Freundlich* isotherm appears to produce a reasonable metal ions on the model for the sorption of the five metals, with Cu(II) seeming to fit the data better than Cd(II), Co(II), Ni(II) and Pb(II). The K_F value of the divalent metal ions followed the order: $\text{Pb(II)} > \text{Cu(II)} > \text{Cd(II)} > \text{Ni(II)} > \text{Co(II)}$, suggesting that Pb(II) has a greater adsorption tendency towards the yam tuber peel waste biomass than the other four metals. The *Freundlich* equation parameter $1/n$, which is a measure of the adsorption intensity and follow the order: $\text{Co(II)} > \text{Ni(II)} > \text{Cd(II)} > \text{Cu(II)} > \text{Pb(II)}$, indicating a preferential sorption of Co(II) by the yam tuber peel waste biomass.

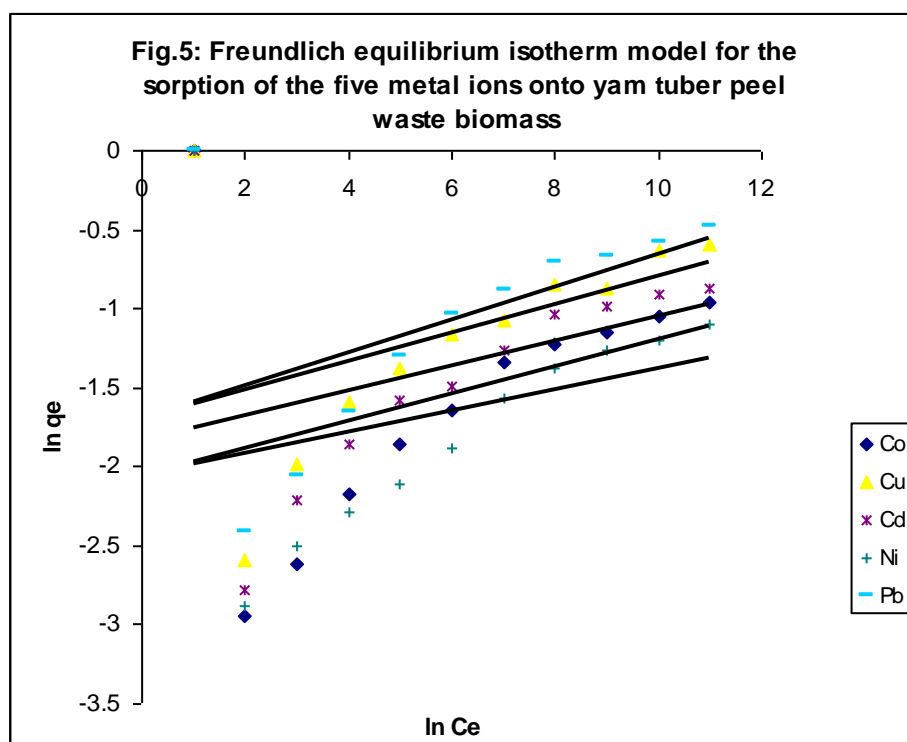


Table 5: Freundlich Isotherm Parameters

Metal ions	1/n	K_F	R^2
Co(II)	0.862	0.012	0.936
Cu(II)	0.693	0.039	0.979
Cd(II)	0.725	0.025	0.975
Ni(II)	0.758	0.015	0.923
Pb(II)	0.575	0.067	0.828

Kinetics of Sorption

Sorption kinetics shows a large dependence on the physical and/or chemical characteristics of the sorbent material, which also influences the sorption process and the mechanism [25].

Pseudo-First Order Model:

A plot of $\ln(q_e - q_t)$ against t (Fig.6) gave the pseudo-first order kinetics. From the plot, it was observed that the *Lagergren* equation did not provide a very good description for the sorption of the five divalent metal ions on *D. rotundata poir* as the coefficient of determination R^2 values (Table 6) are all less than 0.90 except for Ni(II) (0.906). No further consideration of this model was therefore attempted.

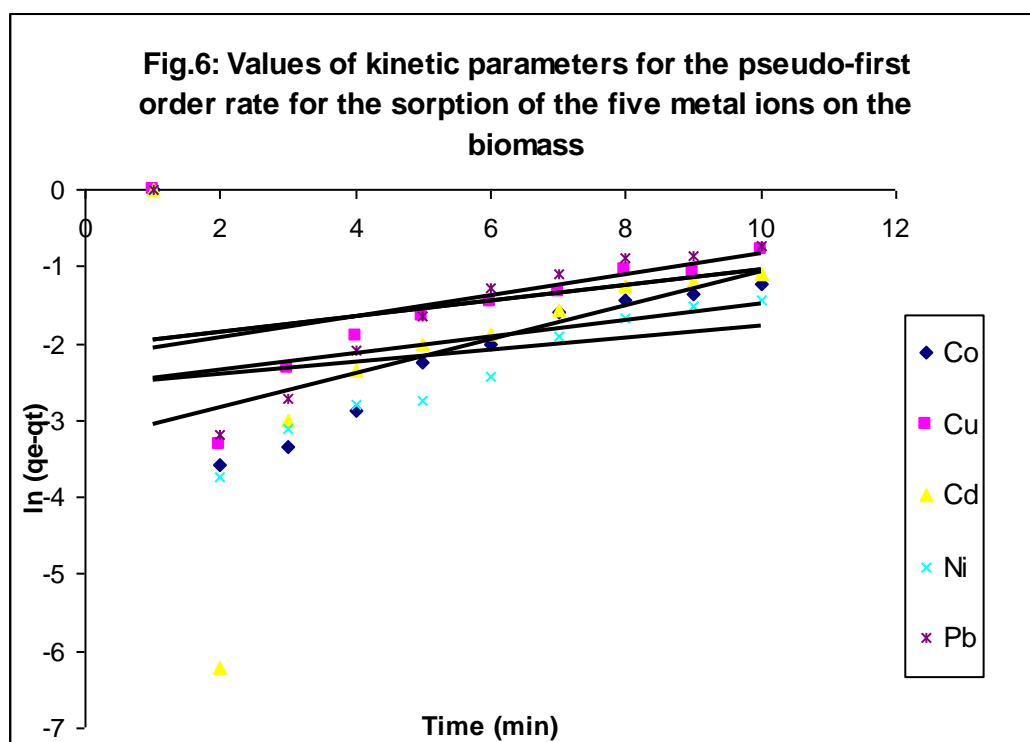


Table 6: Values of Kinetic Parameters for the Pseudo-First Order Rate for the Sorption of the Five Metal Ions on the Biomass

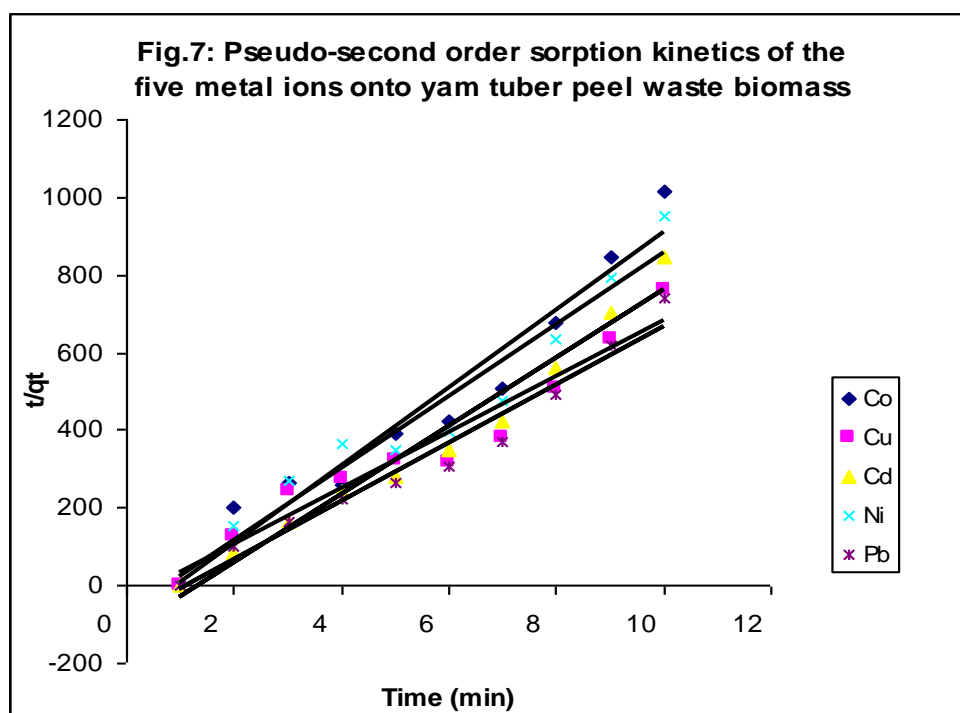
Metal ions	K_1	$q_e(\text{mg/g})$	R^2
Co(II)	0.045	0.033	0.846
Cu(II)	0.037	0.067	0.767
Cd(II)	0.063	0.017	0.552
Ni(II)	0.040	0.030	0.906
Pb(II)	0.042	0.061	0.804

Pseudo-Second Order Model:

The initial sorption rate, h_0 , the equilibrium sorption capacity, q_e , the pseudo-second order rate constant, K_2 , and the coefficients of determination, R^2 , was determined experimentally from the slope and intercept of a plot of t/q_t against t (Fig.7), and are presented in Table 7. The result indicate initial sorption rate follow the order: $\text{Cd(II)} > \text{Pb(II)} > \text{Co(II)} > \text{Cu(II)} > \text{Ni(II)}$. This implies that in a mixed metal ion system of the five metals, Cd(II) may be adsorbed better.

Table 7: Values of Kinetic Parameters for the Pseudo-Second Order Rate for the Sorption of the Five Metal Ions on the Biomass

Metal ions	h_0 ($\text{mg g}^{-1} \text{min}^{-1}$)	K_2 ($\text{g mg}^{-1} \text{min}^{-1}$)	q_e (mg g^{-1})	R^2
Co(II)	0.012	2.840	0.065	0.987
Cu(II)	0.010	1.156	0.093	0.978
Cd(II)	0.050	9.645	0.072	0.999
Ni(II)	0.010	1.876	0.073	0.981
Pb(II)	0.025	3.380	0.086	0.997



Comparism of the coefficients of determination, R^2 for pseudo-first order and pseudo-second order rate laws (Table 8) indicates a good compliance with the pseudo-second order equation, as the coefficients of determination for metal ions on the biomass were >0.988 for the five divalent metal ions. The coefficient of determination for the pseudo-first order kinetic model was smaller than the pseudo-second order, indicating that the pseudo-second order equation is more appropriate in describing the sorption.

Table 8: Comparison of Coefficients of Determination, R^2 , for the Pseudo-First (K_1) and Pseudo-Second (K_2) Order Rate Models.

Metal ions	R^2 from pseudo-first (K_1) order rate model	R^2 from pseudo-second (K_2) order rate model
Co(II)	0.846	0.987
Cu(II)	0.767	0.978
Cd(II)	0.552	0.999
Ni(II)	0.906	0.981

Conclusion

The effects of metal ion concentration on adsorption capacities show that yam (*Dioscorea rotundata* poir) tuber peel waste biomass adsorbed metal ions from solution, with an increase in sorption capacity of the biomass with increased metal ion concentration. The actual percent removal of the metal ions from solution decreased with increase in initial metal ion concentration. The pH profile studies showed that adsorption of the five metals are pH-dependent, with optimum adsorption occurring between pH 4 and 5. This effect suggests that an ion-exchange type processes is responsible for the adsorption mechanism. The adsorption mechanism for these metals is a rapid and stable process and occurred in less than 25min, which implies that adsorption is taking place on the cell wall surface of the yam tuber peel waste biomass. The equilibrium data fitted the *Langmuir* and *Freundlich* isotherms very well. The separation factor or equilibrium parameter obtained from the *Langmuir* isotherm showed that adsorption of metal ions onto the yam tuber peel waste biomass is favourable. The kinetic data clearly establish that pseudo-second order model provides a more appropriate description of the metal ion sorption process of cobalt, copper, cadmium, nickel and lead onto *D. rotundata* poir biomass than a first-order equation. On

the whole, the data showed that, yam tuber peel waste biomass was successful as biosorbent for treating heavy metal contaminated wastewater, and may serve as an alternative adsorbent to conventional means. Hence, not only is *D. rotundata* peel inexpensive and readily available, it also has the potential for metal removal and recovery of metal ions from contaminated waters.

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ICHTNOFABRICS CONTAINING OPHIOMORPHA AND SKOLITHOS IN THE LATE MAASTRICHTIAN GOMBE SANDSTONE, UPPER BENUE TROUGH, NIGERIA: SIGNIFICANCE TO HYDROCARBON EXPLORATION.

O . Odedede

Department of Geology, Delta State University, Abraka,
Email: odededeo@yahoo.com.

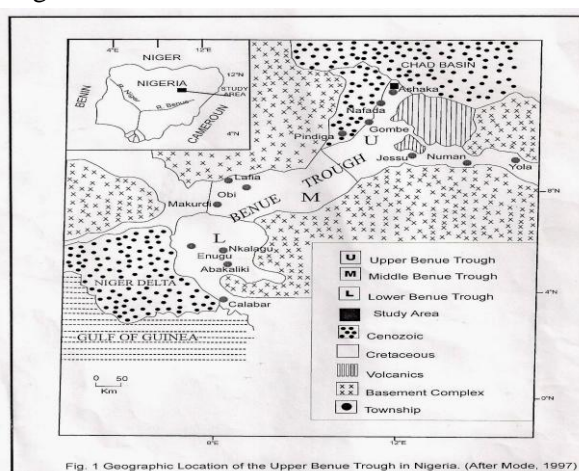
Abstract

The ichnological study of the Late Maastrichtian Gombe Sandstone revealed the existence of marine ichnofaunas in the basin. Bioturbation ichnofabrics display two dwelling burrows; *Ophiomorpha* and *Skolithos*. Sedimentological and ichnological data consistently indicate that deposition occurred in an anoxic to oxygenated bottom water conditions. The bioturbated sandstone facies, fluctuating salinity and ichnological assemblages confirmed marine affinity. Ichnodiversity plot indicate mesohaline to polyhaline waters. High ichnofabric index displayed by *Ophiomorpha* in the bioturbated sandstone alternated with silty clay and favourable sedimentological characteristics of this facies makes it an attractive exploratory target.

Keywords: Ichnofabrics, Bioturbation, Gombe Sandstone, *Ophiomorpha*, *Skolithos*.

Introduction

Ichnofabrics refers to the sediment texture and internal structure arising from bioturbation and bioerosion. Ichnofossils are invaluable palaeoenvironmental indicators (Ekdale, et al., 1984; Frey 1975), have genetic stratigraphic significance and provide additional physical sedimentological data, such as sedimentation rate and event deposition (Gingras et al., 2002), depositional environment and basin analysis (Kowalewski and Demko, 1996) because changes in population structure often reflect environmental signals such as changes in bottom oxygenation, hydrodynamics and salinity etc. Trace fossils also show behavioural responses to changes in sedimentation rate, substrate cohesiveness and sediment grain size. The ichnofabric study of the Gombe Sandstone which outcrops along Wuro Boppar Road in North Eastern Nigeria (Fig. 1) forms the basis of this study. Detailed ichnological studies that attempt to determine palaeosalinity characterization and its implications for hydrocarbon exploration are rare in the basin. However, a few detailed ichnological studies have been published. For example, Mode (1998) studied the distribution and palaeoenvironmental significance of trace fossils in the Late Maastrichtian Gombe Sandstone in the Zambuk area and concluded that it was a sheltered marginal marine bay bounded by an escarpment or uplifted highland.



Geological setting

The Benue Trough of Nigeria is an elongate NE-SW trending intracratonic sedimentary basin, located at a major re-entrant in the West African continental margin. It is subdivided into the Lower Benue Trough, Middle Benue Trough and the Upper Benue Trough. The Upper Benue Trough branches into an E-W trending Yola arm and N-S trending Gongola Basin (Fig. 1). The Cretaceous history of the Upper Benue Trough represents an active rifting followed by a thermotectonic sag stage and tectono-eustatic transgressive and regressive events (Zaborski, 2003). There were mid Santonian and terminal Cretaceous compression leading to uplift of the Cretaceous sediments. However, the Gombe Sandstone Basin appears to have developed as a result of a renewed phase of rifting that occurred during Campano-Maastrichtian times.. Benkhelil (1982), Benkhelil (1986) and Guiraud (1990b) studied the structural geology. Zaborski et al., (1997), Zaborski (2003) and among others investigated the stratigraphy, structure, depositional facies and Cretaceous system in the basin. Although numerous geological studies have been made in the basin, none of these previous workers considered the significance of ichnofabrics in palaeoecology and hydrocarbon exploration.

Materials and Methods of Study

The datasets of this study consists of sedimentological and ichnological data gathered through detailed mapping of the partially exposed outcrops of the Gombe Sandstone. The sedimentary texture, and structures, nature of bedding contacts and trace fossil content were documented. Ichnological observations focused on the identification of ichnogenera, distribution of ichnofossils and assemblages. Trace fossils (tube diameter) and depth of burrows were also measured. Ichnofabric index method was employed to evaluate spatial variability of ichnofabrics within facies. Palaeosalinity diagram of Buatois et al., (1997), Gingras et al., (2002), and Frey (1975) methods of environmental interpretations were also utilized.

Results

Sedimentary facies

On the basis of sedimentological and ichnological data, the Gombe Sandstone display four sedimentary facies, thinly laminated silty shale (Fig. 2), bioturbated sandstone interbedded with silty shale, bioturbated sandstone with silty clay and massive sandstone facies (Table I). The following sedimentary succession occurs south of Biri Bolewa, from top to bottom:

- i. 6 m of massive coarse grained quartz arenite
- ii. 1 m of rapidly alternating silty clay and sandstone. The facies is bioturbated with *Ophiomorpha* burrow traces (Fig. 3) sizes of burrow range from 36– 38 cm in length and 4 – 6 cm in diameter and there are V-in-V laminae at the upper end of burrows (Fig. IV) wedging beds which dip at a low angle are also observed
- iii. 1m of bioturbated sandstone interbedded with silty shale at the base. This facies is intensively bioturbated with *Skolithos* and *Ophiomorpha* burrows. Size of burrows ranges from 9 -20 cm in length and 2-3 cm in diameter.
- iv. 1m of thinly laminated silty shale and lack, bioturbated.



Fig. 2 (colour online): *Skolithos* and *Ophiomorpha* Burrows in the Gombe Sandstone (2km West of Biri Fulani)

Table 1: Summary of Sedimentological, ichnological, and palaeoecological interpretations in the Gombe Sandstone Around Biri Bolewa.

Facies	Ichnofabric	Ichnofabric index	Occurrence and contacts	Sedimentology	Ichnology	Palaeoecological and Palaeo-environmental interpretations
1.Laminated silty shale.	Unbioturbated	0	<ul style="list-style-type: none"> Underline bioturbated sandstone + silty shale Transitional. 	<ul style="list-style-type: none"> Parallel laminated (thinly) Lack of bioturbation 	<ul style="list-style-type: none"> Trace fossils are absent. 	<ul style="list-style-type: none"> Low hydraulic energy Deposited from suspension . Stressed environment due to salinity fluctuation . Anoxic
2.Bioturbated sandstone + silty shale.	<i>Ophiomorpha</i> and <i>Skolithos</i>	0.4	<ul style="list-style-type: none"> Overlie silty shale facies. Gradational. 	<ul style="list-style-type: none"> Parallel bedded and laminated. Compose of quartz arenite at the top . Intensively bioturbated. 	Completely bioturbated. Ichnogera observed include <i>Ophiomorpha</i> and <i>Skolithos</i> burrows.	<ul style="list-style-type: none"> Marine setting. Well oxygenated. High hydraulic energy. Mesohaline to Polyhaline.
3.Bioturbated sandstone +silty clay facies.	<i>Ophiomorpha</i>	0.6	<ul style="list-style-type: none"> Underline massive sandstone facies. Gradational 	<ul style="list-style-type: none"> Intensively bioturbated. Parallel bedded and laminate. High sedimentation rate and shifting substrates. 	<ul style="list-style-type: none"> Completely bioturbated. Ichnogenera (<i>Ophiomorpha</i>) 	<ul style="list-style-type: none"> Modes of sediment transport is by saltation Mesohaline-polyhaline Well oxygenated. Nearshore setting.
4.Massive sandstone	unbioturbated	0	<ul style="list-style-type: none"> Overlie bioturbated sandstone + silty clay facies. Transitional 	<ul style="list-style-type: none"> Medium to coarse grained. Moderately well sorted, slightly micaceous 	<ul style="list-style-type: none"> Absent of trace fossils 	<ul style="list-style-type: none"> High energy setting. Deposition from saltation. Lack of ichnofauna probably due to low oxygen content and salinity stress.

Notable observation recorded around the present study area, is the lack of bioturbation at the lower and upper parts respectively. This may reflect fluctuating salinities, intolerable bottom conditions and rapid accumulations of sediments over the burrowing organism as evidenced by the termination of the burrows at the massive sandstone (Fig 3). Massive beds deposited in this condition mostly arise through rapid sedimentation (Odedede, 2002). The lower unit is interpreted to have been deposited within a transition to lower shore face setting as evidence by the lithofacies, while the upper part indicate littoral-shallow neritic environments.

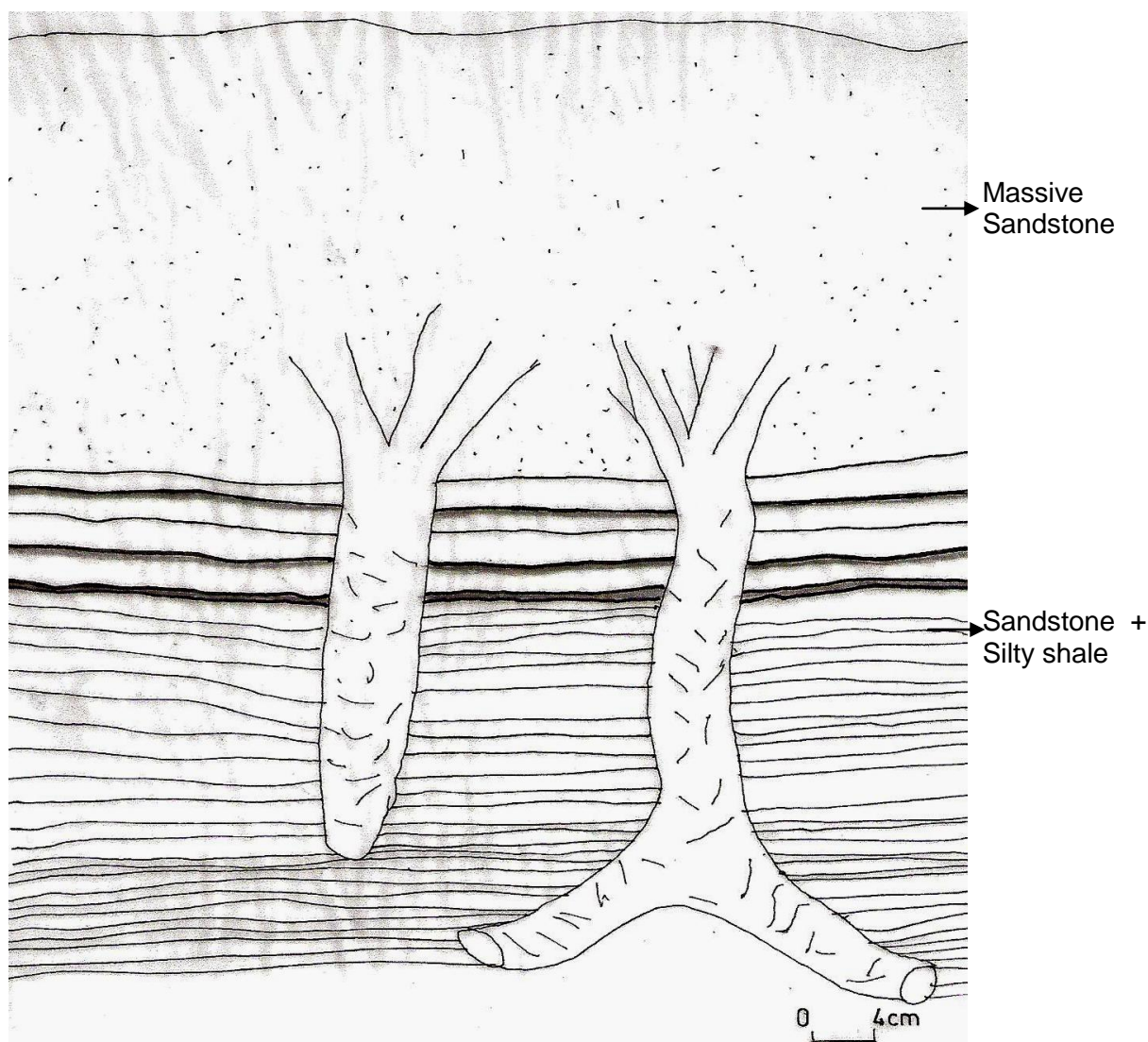


Fig. 3: Feeding traces made by *Ophiomorpha Sp.* In Gombe Sandstone with associated “Chevron Structure” and truncated at the massive sandstone, along Wawa-Wuro Boppar Road 2 ½ km West of Biri Bolewa, Gombe State, Nigeria.

Palaeoecological Reconstruction

Trace fossil diversity differs from animal species diversity (Bromley, 1996) and provide some basic information on trends in species richness in marginal marine environments. Two species of ichnofaunas plotted on number of species versus salinity gradient diagram of Buatois et al., (1997) suggest salinity ranges from mesohaline to polyhaline (Fig. 4). The diversity of freshwater animals tends to decline rapidly as salinity increases, but diversity of marine organisms decreases (Fig. 4) more gradually with the dilution of normal marine salinity (Benyon and Pemberton, 1992; Buatois et al., 1997). Though, the ichnodiversity recorded in this area is low, sedimentologic data, presence of lateral and vertical

distribution of ichnotaxa indicate a regime of anoxic to oxygenated water conditions and fluctuating salinity (Fig. 3). These scenario may account for the lack of biogenic structure and low ichnodiversity encountered at the bottom of the Gombe Sandstone section.

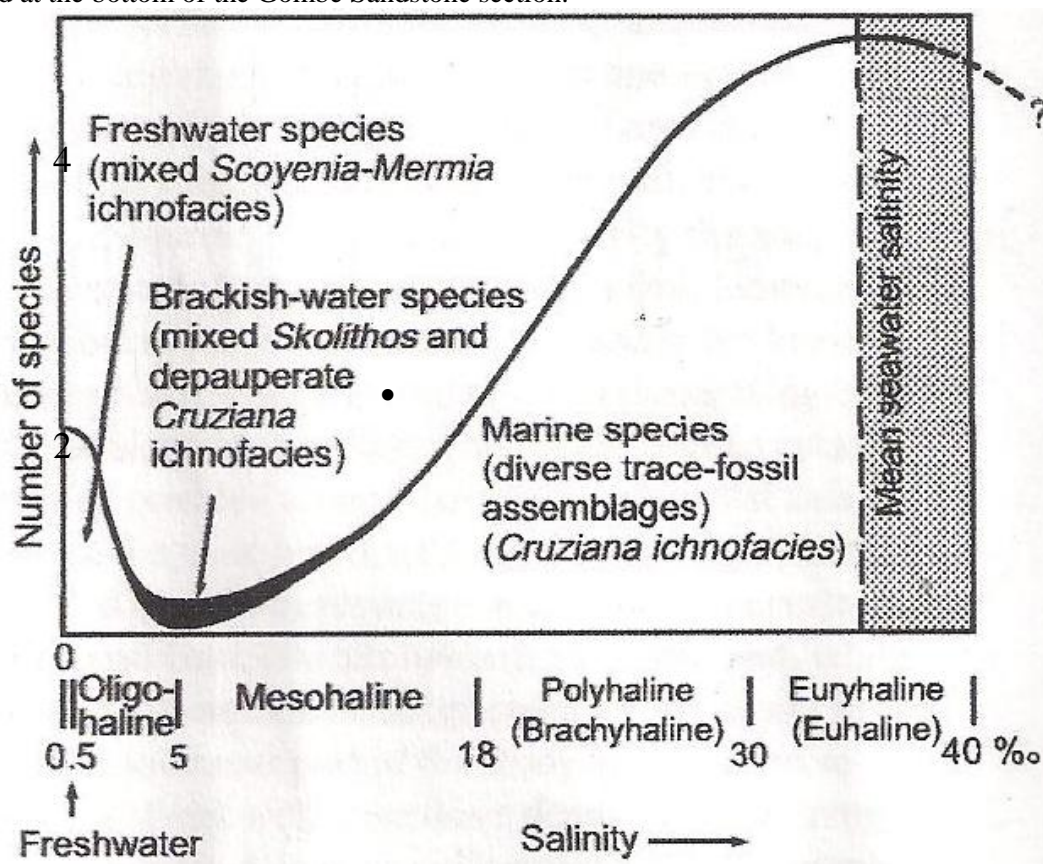


Fig. 4: Relationships among species diversity, ichnofaunas, and salinity along a salinity gradient (Modified after Buatois et al., 1997).

Discussion

Ichnofabrics encountered in the study area are essentially dwelling traces (*Skolithos* and *Ophiomorpha*). The *skolithos* burrows are straight to slightly curved, thinly lined and U-shaped occurring perpendicular to bedding surface (Fig. 3). One of the now-classic littoral and sublittoral sand body trace fossils is *Ophiomorpha* (Weimer and Hoyt, 1964). In recent sediments such structures are produced by a variety of crustaceans, the best known being the ghost shrimp *Callinasa major* (Weimer and Hoyt, 1964). The animal observed primarily in the low littoral zone between mean sea level and low tide on beaches that face the open ocean by Weimer and Hoyt (1964). *Ophiomorpha* thrives whenever salinities and current energy are moderately high and the substrate consists of mainly sand (Howard and Frey, 1973); some *Ophiomorpha* also prefer muddier substrates in waters of slightly less current energy. The latter is consistent with the present study as evidenced by the occurrence of *Ophiomorpha* in the rapidly alternating silty clay and sandstone (Fig. 3) and the truncated burrows at the massive sandstones (Fig. 3). Another important feature observed, is the V-in-V laminae associated with the *Ophiomorpha* burrow (Fig. 3). These structures may be as a result of burrow abandonment collapse and the settling downward of superjacent sediment (Odedede, 2002). These ichnofabric associated with fine-medium grained, well bedded quartz arenite and rapidly alternating silty clay, and sandstone interbedded with silty shale are interpreted as representing shallow marine deposits and are consistent with those described from the littoral and shallow neritic environments (Weimer and Hoyt, 1964; Frey, 1975).

Finally, ichnofabrics produced in the bioturbated sandstone interbedded with silty shale facies display a low diversity, presence of primary sedimentary structures and dwelling burrows correspond to

near shore setting, while the upper facies with more abundant laminations, increase in suspension feeding structures, alternating with the deposit feeding structures were consistent with a moderate energy setting. This facies grades upward into a massive quartz arenite, unbioturbated and weakly storm influenced environment is interpreted, based on the coarsening upward character of the succession. The thicker and greater number of storm beds corresponds to progressive shallowing along the shoreface depositional profile (MacEachern and Pemberton, 1992; MacEachern et al., 1998). The upward increase in grain size, sedimentary structures, and sandstone content also confirmed the shallowing of the basin during fill. The trace fossils suite displays strong evidence of anoxic condition and environmental stress near the base of the succession, due to lack of bioturbation, but is progressively less stressed near the top of the succession (Fig. 2), particularly as food quality increases, salinity content and a change from anoxic to oxygenated conditions resulting to high ichnofabric index recorded in this lithofacies. These findings strongly support the earlier reports of Zaborski et al., (1997), and Odedede (2002) that suggested shallow marine origin for the bedded sandstone facies.

Implications For Hydrocarbon Exploration .

Ichnofabrics analyses have profound implications for hydrocarbon exploration because reservoir quality is largely influenced by external geometry and distribution of depositional facies. Sedimentologic and ichnologic analyses of the Gombe Sandstone indicate a high variability in sedimentary facies controls reservoir heterogeneities. Bioturbated quartz arenite with rapidly alternating silty clay facies may have a good quality reservoir sands than the bioturbated quartz arenite with silty shale (baffles flow) facies because the silty clay may act as seals and restrict the flow of fluids while the sandstone units exhibit reservoir attributes. Generally, assumption is that bioturbation reduces porosity and permeability, but this work contrast this assertion. According to Gingras et al., (2002) that deposit feeders that backfill their burrows may damage pore connectivity in certain situations, but open structure (Fig. 3) produced by suspension feeders and passive carnivores do not reduce porosity and permeability and may even act as conduits for fluid migration (Fig. 3). The present study lends support to this interpretation. In addition, high ichnofabric index displayed by *Ophiomorpha* (Table I), suggest an attractive reservoirs. According to Anderson and Droser (2002) within a sequence stratigraphic context reported that bioturbations by *Ophiomorpha* are more pervasive in lowstand systems tract compared to transgressive systems tracts. This is consistent with the predominance of marginal and near shore marine sand dominated settings that are characteristic of lowstands, which are favourable habitats for colonization by *Ophiomorpha* producers. The predominance of *Ophiomorpha* in the bioturbated facies (Table 1) may point to its good reservoir attributes. Also the high ichnofabric index (0.6) and favourable sedimentological characteristics of this facies makes it an attractive exploratory target.

Conclusion

The partially exposed Gombe Sandstone around Biri Bolewa, Gombe State, NE, Nigeria, consists of silty shale, bioturbated sandstone interbedded with silty shale, bioturbated sandstone with rapidly alternating silty clay and massive quartz arenite. Ichnofabric study revealed the existence of dwelling traces of marine ichnofaunas; *Skolithos* and *Ophiomorpha*. Sedimentological and ichnological analysis suggest a regime of anoxic to oxygenated bottom water conditions, mesohaline to polyhaline waters and high ichnofabric index display by *Ophiomorpha* in the bioturbated sandstone adjudged this facies as a good indicator of lowstand system tracts, which are attractive exploratory targets. Sedimentological and ichnological analysis undertaken in this study may be applied to subsurface outcrops in new exploration programmes, so as to reduce prospect risk and enhance production strategy.

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INVESTIGATION OF SAND DEPOSIT IN PARTS OF OKPE LGA , DELTA STATE, NIGERIA.

¹Atakpo E. A. and ²Akpoborie A. I.

¹Department of Physics, Delta State University, Abraka.

²Department of Geology, Delta State University, Abraka.

¹Email: e_atakpo@yahoo.com

Abstract

The exploration and exploitation of sand an important industrial and local raw material with a wide application and uses has been on the increase in Delta State, Nigeria. Geoelectric investigation for sand deposit was carried out in parts of Okpe local government area using 2D and 1D Electrical Resistivity Sounding. Three distinct layers comprising topsoil, laterite/Clayey sand and sand were delineated from the 2D and 1D interpretation. The topsoil has thickness varying from about 0.8 to 1.5m while its resistivity values vary from 195 to 2843 Ω m. The lateritic/clayey sand layer has resistivity values ranging from 192 to 2808 Ω m and thickness of about 5 to 8m. The thickness of the sand layer varies from about 5 to 40m while the resistivity values ranges about 1119 to 8500 Ω m. This layer consisting of fine grained sand constitutes the sand deposit. The study has shown that the area is very viable for sand exploitation.

Key Words: Sand, Geoelectric, Delta State

INTRODUCTION

Sand is one of the industrial and local raw materials available to mankind with a wide range of application and uses such as the production of glass, construction of buildings and roads. The investigation of sand deposits is necessary because of its importance to economic development.

Economic sand deposits in the Niger Delta have been reviewed by Akpokodje and Etu-Efeotor (1987), and possible glass sand deposits have been studied in the eastern delta by Ushie, Esu And Udom (2005). The economic potential of the sands is also reviewed in the Delta state, Nigeria, Economic Atlas (2009), also by the Delta state Ministry of Commerce and industry (2001). Glass sands are also being exploited at the glass factory in Ughelli.

Beyond this, sand deposits are currently mined in Delta State through so called borrow pits: large, unregulated and haphazardly located excavations. These traditional artisanal operations are hardly preceded by any formal exploratory studies or assays such that the economic worth or futures of a specific location is uncertain. In order to be included in the mainstream of formal economic planning and for the exploitation of these obviously important deposits to be controlled by appropriate guidelines, the deposits need to be properly mapped for eventual parceling into leases for licensing purposes as is the case with other industrial mineral deposits.

The geoelectric method can be used for the rapid and detailed investigation of sand deposits. The success of employing the geoelectric method has been found to depend on proper acquisition and interpretation of obtained data, which requires a careful correlation of the geophysical data collection to known geology of the area (Beck, 1981).

This method involves the measurement of apparent resistivity of subsurface materials as a function of depth or position. It has been used successfully for subsurface investigation for clay, gravel, kaolin deposits and ground water investigation (Iserhien-Emekeme et. al., 2007, Ananaba et. al., 1993 Afolabi et al. 2004 and Atakpo and Akpoborie, 2008).

In the present study, the 2D geoelectric method using the Dipole-Dipole array and the 1D geoelectric method using the Schlumberger array have been employed to determine the nature of the superficial material and the subsurface rocks underlying it with a view of locating sands deposits of commercial value.

LOCATION AND GEOLOGY OF STUDY AREA

The study area lies within longitude $5^{\circ}50'01''$ and $5^{\circ}50'35''$ East and Latitude $5^{\circ}35'32''$ and $5^{\circ}35'34''$ North (Figure 1) in the low lying Sombreiro-Warri Deltaic Plain of the Western Niger Delta. This coastal plain is situated in the tropical rain forest region. Records obtained from the Nigerian Meteorological Agency in Warri showed the ten year mean (1980-1991) annual rainfall, annual temperature and humidity of 2700mm, 27.23C and 78.73% respectively.

The Sombreiro - Deltaic plain is a physiographic feature built by delta top distributary lacustrine and fluvial processes that continue till present day (Allen, 1967). It is thus filled by a Quaternary-Recent alternating sequence of silts, medium to coarse grained sands, sandy clays and discontinuous clay bands that mask the Benin Formation. While these deposits are thought to be recent expressions of and continuation of present day deposition of the Benin Formation, they possess distinct hydraulic and other engineering characteristics (Bam, 2007) that are important for water supply as well as other purposes.

The Benin Formation is the youngest of the three formations that fill the Niger Delta Basin. The other formations are the paralic sand and shale Agbada Formation (Eocene to Oligocene) and the basal Akata Formation (Palaeocene to Eocene) that comprises mainly of marine shales and sand beds. The approximately 2000m thick massive Benin Formation contains friable sandy beds that are exploited extensively for construction purposes. The depth to water in the The Sombreiro - Deltaic plain is at a maximum of 5 m below the ground surface in the dry season and less than 1m in the wet season (Akpoborie et al., 2000).

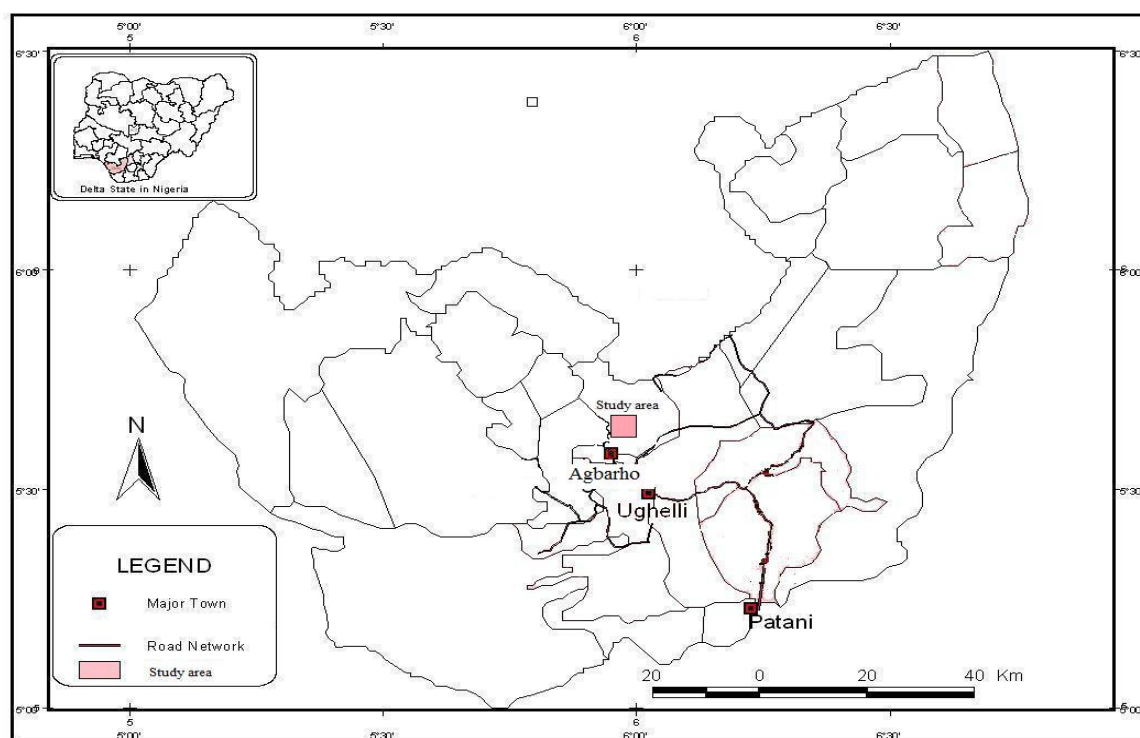
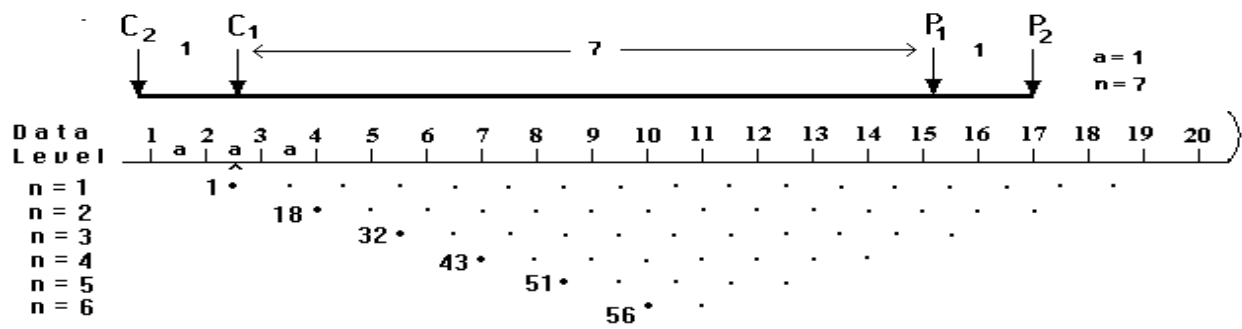


Fig. 1 (colour online): Location of Study area

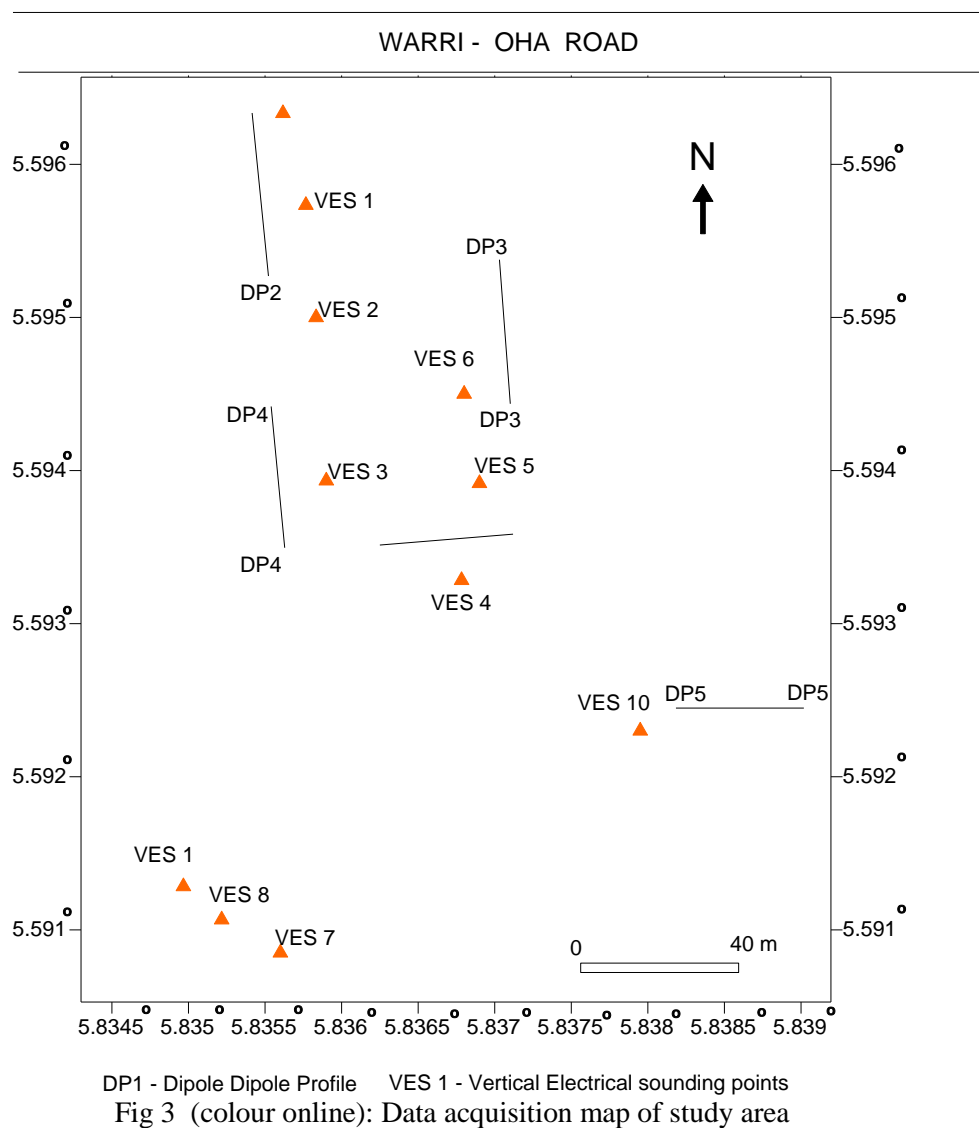
FIELD DATA ACQUISITION

The 2D survey was carried out using the SAS 1000 Terrameter. 2D surveys are usually carried out using a large number of electrodes measurement positions (Fig. 2).



Sequence of measurements to build up a pseudosection

Fig. 2. The arrangement of electrodes for a 2D electrical survey and the sequence of measurements used to build up a pseudosection.



The dipole- dipole array method was adopted for the 2D resistivity survey. A total of five profiles were occupied in the study area with maximum distance ranging from 75 to 160m (Fig.3). Electrode separation

of 10m between adjacent electrodes was used for profiles (1-4) while a separation of 5m was used for profile 5. The stored data in the Terrameter was transferred to a computer for processing and inversion using the DIPPRO inversion software. In addition to the 2D investigation 1D resistivity sounding also was carried out. Ten (10) vertical electrical sounding by adopting the Schlumberger method was carried out at preferred points in the study area (Fig.3). The ABEM SAS 1000 Terrameter was also used for the 1D resistivity measurements. The maximum current electrode separation was 500m. The large electrode spread was aimed at achieving greater depth of exploration. The data obtained from the 1D electrical resistivity survey was plotted on a log-log graph paper with the electrode separation ($AB/2$) on the abscissa and apparent resistivity (ρ_a) values as the ordinate. The true resistivity and thickness of the subsurface layers were interpreted by partial curve matching with the two layer model master curves and the corresponding auxiliary curves. The thickness and resistivity values obtained from the partial curve matching were then used for a quantitative computer iteration using the Resist Software based on the work of Vander Velpen (1988).

Ugolo1 (2-D Resistivity Structure)

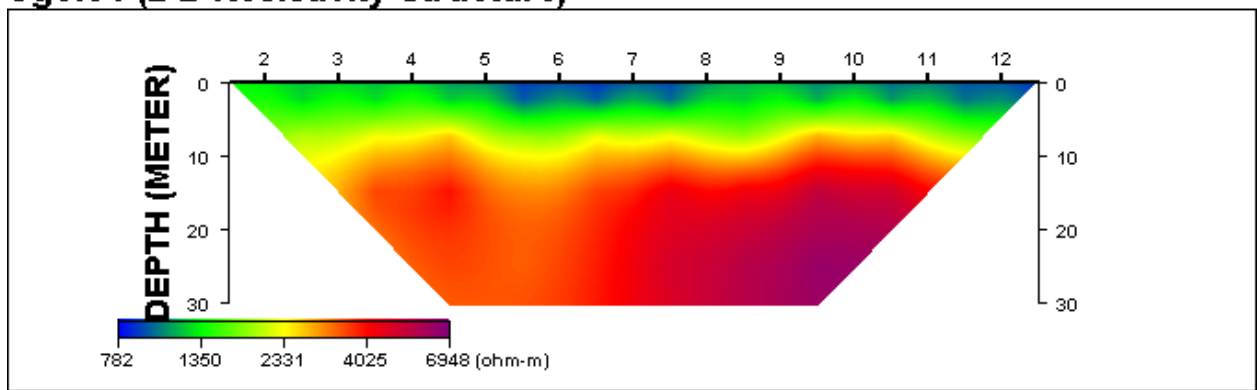


Fig 4a (colour online) : 2D Resistivity Structure of profile 1

UGOLO2 (2-D Resistivity Structure)

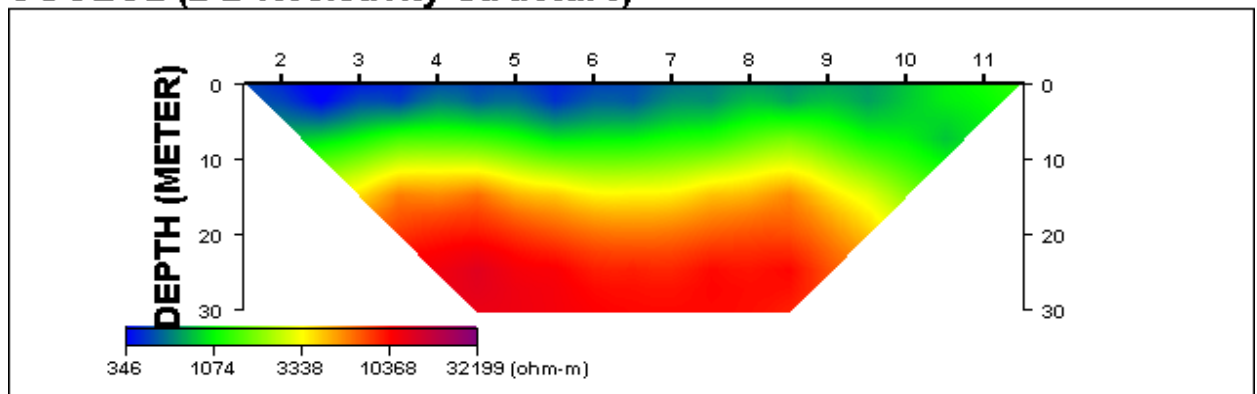


Fig 4b (colour online): 2D Resistivity Structure of profile 2

Ugolo3 (2-D Resistivity Structure)

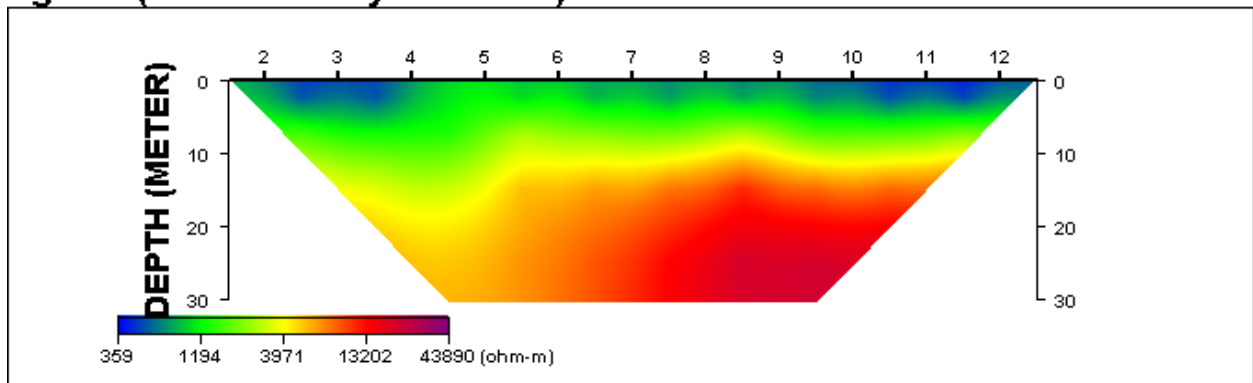


Fig 4c (colour online): 2D Resistivity Structure of profile 3

Ugolo4 (2-D Resistivity Structure)

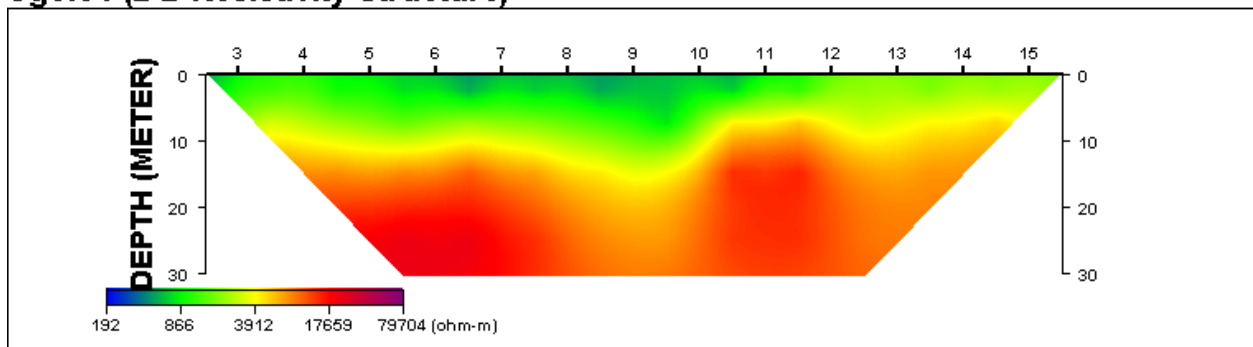


Fig 4d (colour online): 2D Resistivity Structure of profile 4

Ugolo5 (2-D Resistivity Structure)

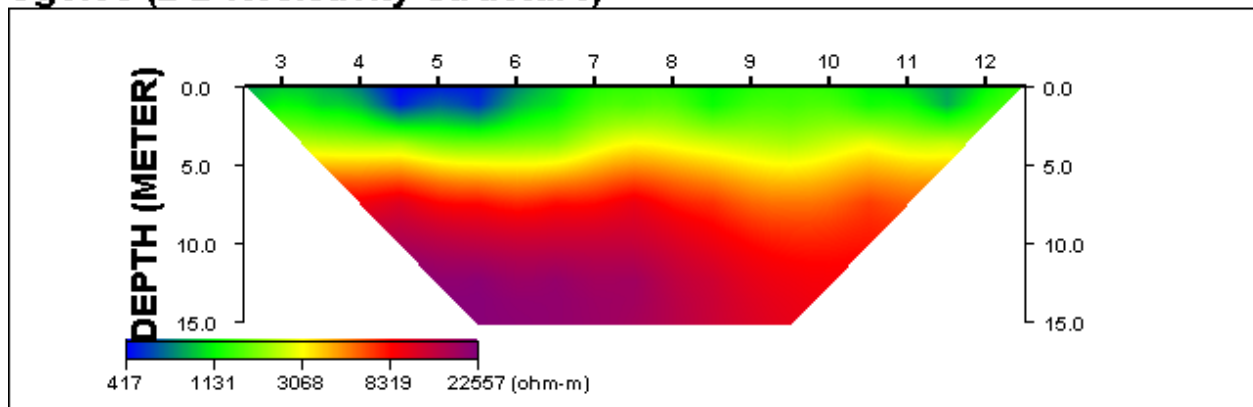


Fig 4e (colour online): 2D Resistivity Structure of profile 5

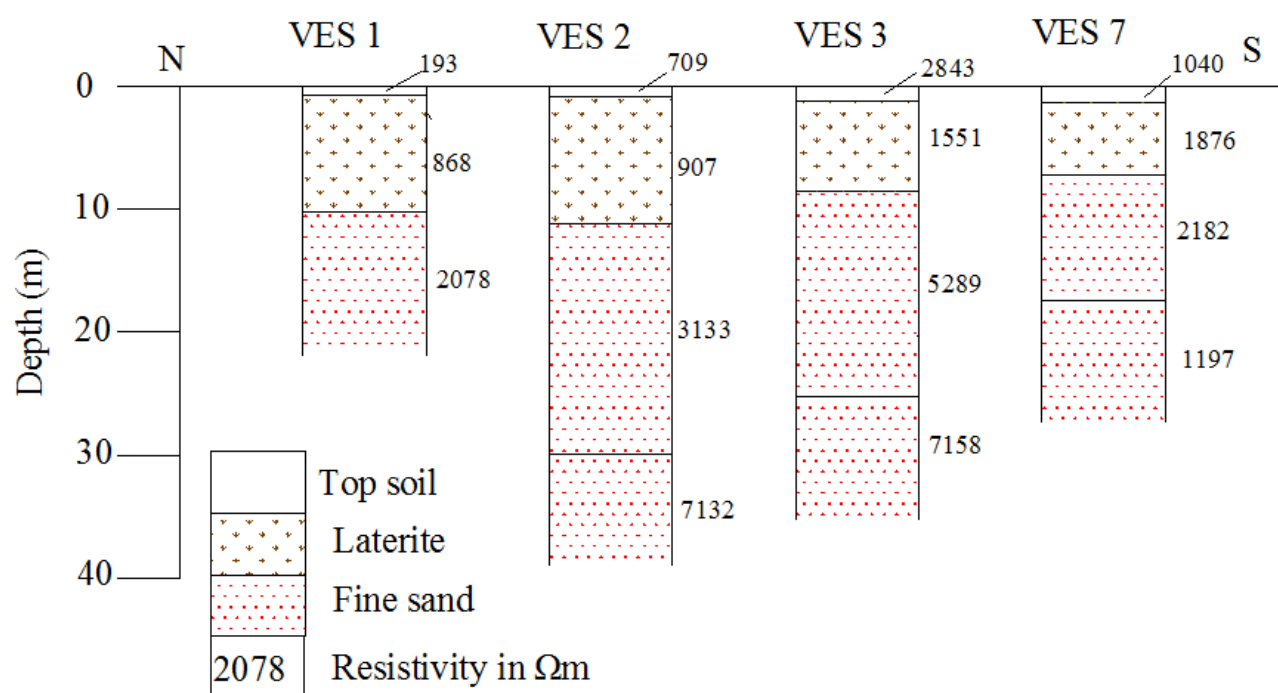


Fig 5a (colour online): Geoelectric section of study area

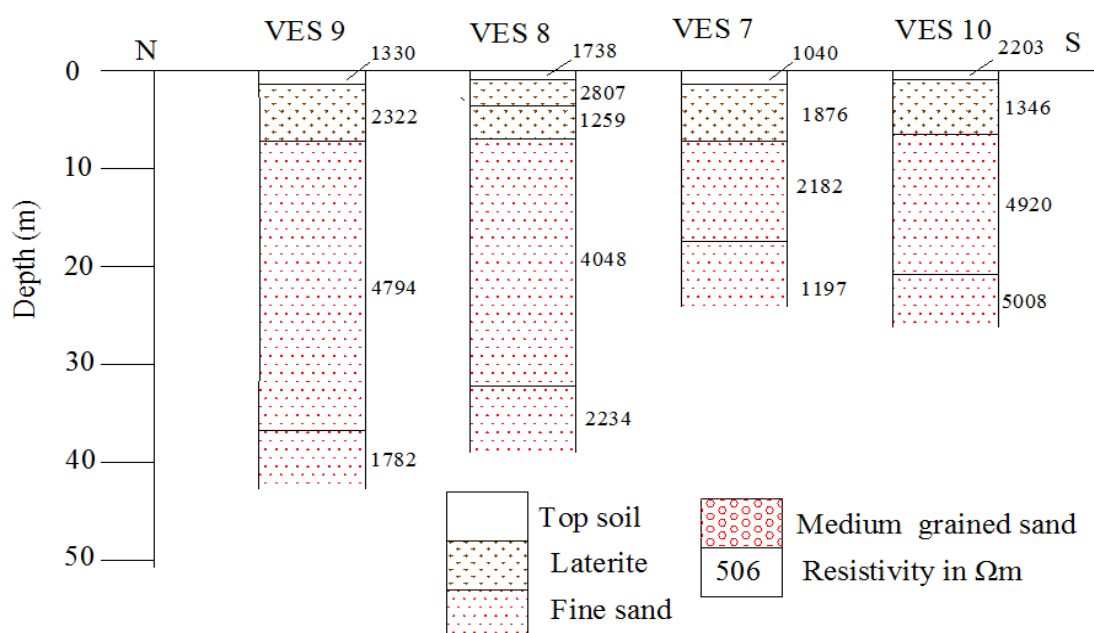


Fig 5b (colour online): Geoelectric section of study area

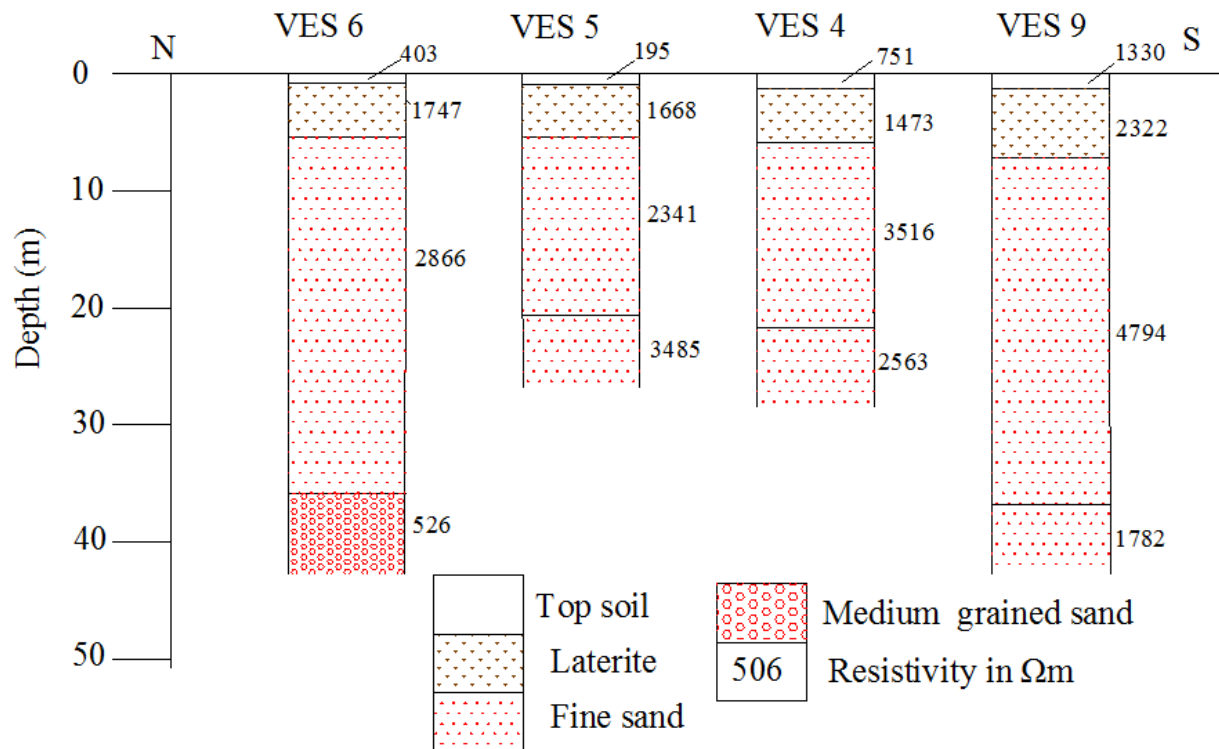


Fig 5c (colour online): Goelectric section of study area

RESULTS

The results of the 2D resistivity inversion are shown in Figs. 4a to 4e while Figs 5a to 5c are the the 1D inversion results.

Fig 4a is the 2D Resistivity Structure of profile 1. The inverted resistivity model showed variation of resistivity values ranging from about 346 to 5200 Ωm . The top soil has resistivity values ranging from about 700 to 1200 Ωm and the thickness of about 1.0 to 2m. The second geoelectric layer has resistivity values varying from 1000 to 1500 Ωm . The thickness vary from 3.0 – 5m, this layer is lateritic. The third layer which is composed of fine grained sand has resistivity values ranging about 1500 to 3500 Ωm with thickness ranging from 5-10m. The fourth layer also consists of fine grained sand with resistivity values ranging from 3500 to 5200 Ωm .

Fig 4b is the 2D Resistivity Structure of profile 2. The inverted resistivity model showed variation of resistivity values ranging from about 364 to 6000 Ωm . Three distinct layers are revealed along this traverse. The top lateritic layer with resistivity values ranging from 346 to 1074 Ωm and the thickness of about 5 to 10m. The second geoelectric layer has resistivity values varying from 1000 to 1500 Ωm . The thickness vary from 8-20m, while the third layer has resistivity values ranging about 3338 to 6000 Ωm with thickness ranging from 5-10m. The second and third layers consist of fine grained sand

Fig 4c is the 2D Resistivity Structure of profile 3. The inverted resistivity model showed variation of resistivity values ranging from about 359 to 8500 Ωm . Three distinct layers are revealed along this traverse. The top lateritic layer with resistivity values ranging from 346 to 1195 Ωm and the thickness of about 5 to 6m. The second geoelectric layer has resistivity values varying from 1194 to 3971 Ωm . The thickness vary from 5 - 18m, while the third layer has resistivity values ranging about 3971 to 8500 Ωm with thickness ranging from 5-10m. The second and third layers consist of fine grained sand

Fig 4d is the 2D Resistivity Structure of profile. The inverted resistivity model showed variation of resistivity values ranging from about 192 to 6000 Ωm . Three distinct layers are revealed along this traverse. The top lateritic layer with resistivity values ranging from 192 to 866 Ωm and the thickness of about 2-5m. The second geoelectric layer has resistivity values varying from 861 to 2000 Ωm . The thickness varies from

6-10m, while the third layer has resistivity values ranging about 2000 to Ωm 5800 with thickness ranging from 5-10m. The second and third layers consist of fine grained sand.

Fig 4e is the 2D Resistivity Structure of profile 5. The inverted resistivity model showed variation of resistivity values ranging from about 417 to 5600 Ωm . Three distinct layers are revealed along this traverse. The top lateritic layer with resistivity values ranging from 417 to 1002 and the thickness of about 2 -5m. The second geoelectric layer has resistivity values varying from 1100 to 2500 Ωm . The thickness varies from 5-10m, while the third layer has resistivity values ranging about 2500 to 6000 Ωm with thickness ranging from 5-10m. The second and third layers consist of fine grained sand.

DISCUSSION

The geoelectric section of the study area revealed three distinct geoelectric layers namely, topsoil, laterite and sand. The topsoil has thicknesses varying from about 0.8 to 1.5m while its resistivity values vary from 195 to 2843 Ωm . The second geoelectric layer is composed of laterite. The resistivity value of this layer ranges from 868 to 2808 Ωm and thickness of 6 to 10 m. The third geoelectric layer is composed of fine grained sand with resistivity 1197 to 7158 Ωm . The thickness of the sand layer is over 30 m in some VES point. Medium grain sand was encountered beneath VES 6 at 37m depth.

This study has demonstrated the utility of the VES method, 1D and 2D imagery for the rapid, non-invasive and efficient evaluation of possible sand quarry sites. In the process, typical resistivity values for clean and uncontaminated pristine Sombreiro –Warri Deltaic Plain sands in this pollution prone region have been determined. Furthermore, it appears that grain size is increasing with depth and there are no clay bands in evidence at depths penetrated thus further confirming the lensoid nature of the clays that are known to constitute part of the sedimentary fill. This is interesting with respect to the vulnerability of the ground water to contamination because water table conditions are prevalent and depth to water ranges from less than 1m in the wet season to about 5m in the dry season. In the absence of regulations many abandoned sand quarry sites in this region tend to be used as waste dump sites as well as becoming receptacles of storm water runoff thereby constituting sources of persistent groundwater contamination, a real problem in this area (Abimbola, 2002; Egbo et al.,2000).

CONCLUSION

Geophysical investigation has been carried out in the study area using the 2D and 1D geoelectric method. Three subsurface geoelectric layers were delineated namely the topsoil, laterite and sand. The 2D inverted sections and the 1D geoelectric sections have revealed sand deposits from depths of 6 to 30m and may probably extend beyond the area investigated. Sand of commercial quantity can be mined in the study area. The absence of clay is an indication that the area is highly vulnerable to ground water contamination and sand mining should be guided by appropriate regulations that protect the environment.

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INVESTIGATING THE CHARACTERISTICS OF ATLANTIC OCEAN BY STANDARD METHODS AT OGULAHAI IN DELTA STATE, NIGERIA

Umudi E. Q, Awatefe K. J., Okoh B. E. and Odjighere M. O.

CHEMISTRY DEPARTMENT, COLLEGE OF EDUCATION, AGBOR., DELTA STATE.

e-mail: ese.umudi@yahoo.com; Phone : 07032332579

Abstract

The physico-chemical and microbial analyses of water in Ogulhai community in Bomadi Local Government Area of Delta State were carried out by standard methods for water and effluents analysis. The results range as follows: colour (9.20 – 11.30) Hazen units, suspended solids (40.01 – 50.5)mg/L, Total Dissolved Solids (23,000 – 26,001)mg/L, Turbidity (66.00 – 78.00) mg/L, pH (7.6 – 7.8) mg/L, Dissolved Oxygen (4.20 – 4.70) mg/L, Biological Oxygen Demand (2.80 – 3.01) mg/L, Salinity (35.00 – 35.60)%, Sulphate (471 – 480) mg/L, Acidity (8.50 – 10.30) mg/L, Alkalinity (40.00 – 43.00) mg/L, Chloride (1200 – 1299.80) mg/L, Conductivity (29300 – 37700) $\mu\Omega/\text{m}$, Phosphate (0.269 – 0.321) mg/L, Nitrate (0.13 – 0.18) mg/L, Ammonium – Nitrogen (0.13 – 0.21) mg/L, Carbonates (26.00 – 28.00), Bacterial count 4.2×10^2 . These are above World Health Organisation maximum limits except for Alkalinity, which is below. Heavy metal concentrations determined were also above World Health Organisation (WHO), Standard Organisation of Nigeria (SON) and Federal Environmental Protection Agency (FEPA); Lead, Chromium and Mercury were below detectable limit. The results show that the water in Ogulhai community fails to meet WHO Standard for potable water.

Keywords: Physico-chemical, Ogulhai, portable.

INTRODUCTION

The importance of water to human survival on earth is highlighted by Ademoroti, (1996). He held that human activities and settlement hinges on the availability of water. Water is vital to life. Hence any water supply in desired quantity and quality is treated with seriousness in most part of the world (El-Nakhai, 2002). The problem of insufficient potable water is more pronounced in the wetland or creeks because of the terrain. Government could scarcely afford the cost of infrastructural outlay needed to provide potable water for her citizens. The problem is further compounded by limited technology, insufficient technical inputs, poor maintenance culture (for few existing water facilities), and no requisite skills. (Okoye, and Adeleke, 1991).

For those living in the Ogulhai community they depend on this ocean water or that provided by Shell Oil Company – which is only available at their own time. At Ogulhai water supply source provided by the Niger Delta Development Commission is not functioning. The Ogulhai people depend on almost solely seawater for their daily need. There is the need to analyse some of the physical, chemical and bacteriological characteristics of the sea water. This work therefore examined some water quality parameters of the seawater as a portable water source for the inhabitants.

Study Area

Ogulahai is a community in Bomadi Local Government Area of Delta State endowed with biodiversity (Ejemeyovwi, 2006). It is located between Latitudes $5^{\circ} 35'N$ and $5^{\circ} 49'N$ and Longitude $5^{\circ} 46'E$ and $5^{\circ} 49'N$. (Ejemeyovwi, 2006). It lies in the mangrove belt of Nigeria, with swamp forest occurring in flat floored valley and adjoining low-lying areas that are seasonably or permanently waterlogged. The soils are deeply weathered and nutrient deficient. The climate is humid subequatorial. It rains almost throughout the year, with temperature of about $27 - 30^{\circ}C$ with seasonal variation not exceeding $3^{\circ}C$. Humidity is high. It has multiplicity of creeks and rivers bordered by mangrove trees which thrive best in the saline environment.

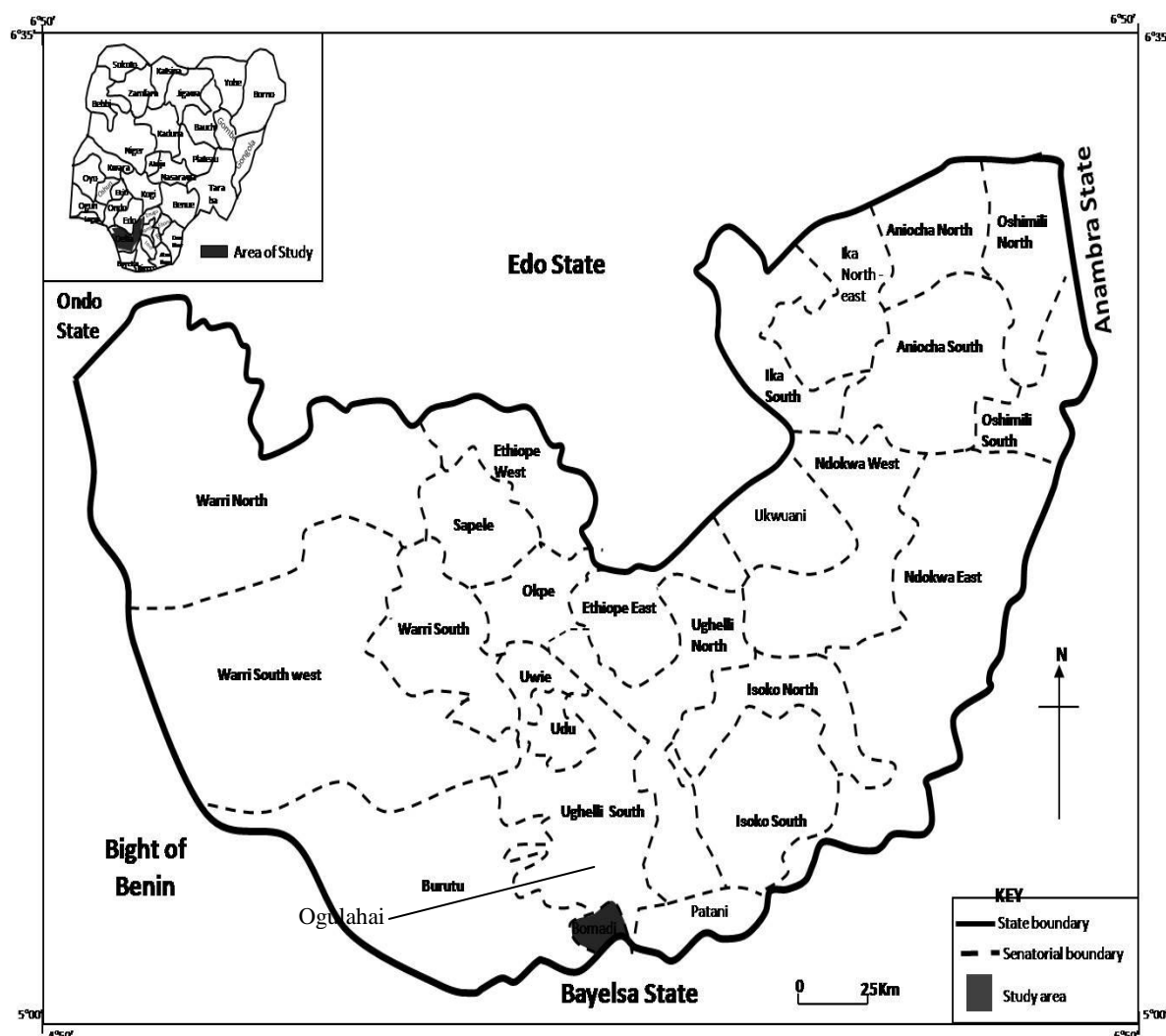


Fig 1 : Map of Delta State Showing Study Area

Source: Modified after Ministry of Lands and Survey, Asaba, 2008

Methodology

Samples were taken between October 2008 and August 2009.

Plastics were treated with dilute chromic acid, water, distilled water and seawater from the sampling source. Ten (10) sampling sites were chosen within distance of 1.2Km. pH determination were done using HACH HQ20 while colour was measured using DR 4000U, turbidity was measured by Nephelometric method using Turbidimeter HACH 2100P. Ademoroti (1996).

Total Dissolved Solids by Photometer method using HACH DR 2010, salinity by Electrical conductivity method using HACH SERISION-5 conductivity meter. While Dissolved Oxygen was by

using Hach DO meter, Biological Oxygen Demand determination by Azide modification method, Chemical Oxygen Demand by close Reflux, Titrimetric method American Public Health Association (APHA) (1992). Acidity, Alkalinity and Carbonates are by titrimetry methods. Phosphates was determined by Ascorbic Acid method using Hach DR4000U. Ammonium – Nitrogen was done by Direct Nesslerization method using Hach 4000U, Nitrate by Cadmium Reduction Method while chloride was done using Argentometric method. American Public Health Association (APHA) (1992). Heavy metals were determined using GBC scientific Atomic Absorption Spectrophotometer (AAS) analyser while sodium and potassium were determined using Bulk Scientific Flame Emission Spectrophotometer (FES)/Flame Absorption and total bacterias using Ademoroti (1996).

Results and Discussion

Table I: Physico-chemical Parameters of Seawater

Parameters/Units	WHO Value (EST DEV)	Wet Season (Mean Value) ± ESTDEV	Dry Season (Mean Value) ± ESTDEV
Colour (Hazen units)	3.00	9.20	11.30
Temperature (°C)	25.00	26.30	28.51
SS (mg/L)	500.00	40.01	50.5
TDS (mg/L)	1000.00	26001.00	23,000.00
Turbidity (mg/L)	5.00	78.00	66
pH	7.90	7.80	7.60
DO (mg/L)	-	4.20 ± 0.10	4.70 ± 0.14
BOD (mg/L)	-	3.01 ± 0.05	2.80 ± 0.05
COD (mg/L)	-	821 ± 1.50	837 ± 1.55
Salinity (‰)	-	35.00 ± 1.60	35.60 ± 1.61
Sulphate (mg/L)	250.00	481.00 ± 1.73	475.00 ± 1.65
Acidity (mg/L)	-	10.30 ± 0.21	8.50 ± 0.19
Alkalinity (mg/L)	100.00	41.00 ± 0.20	43.00 ± 0.21
Chloride (mg/L)	200.00	1200.10 ± 1.08	1299.80 ± 1.31
Conductivity (µΩ/m)	900.00	29300.00 ± 0.13	37700.00 ± 0.21
Phosphate (mg/L)	-	0.321 ± 0.03	0.269 ± 0.03
Nitrate (mg/L)	10.00	0.18 ± 0.02	0.13 ± 0.00
NH ₄ ⁺ -N (mg/L)	-	0.21 ± 0.000	0.13 ± 0.003
Carbonates (mg/L)	100.00	26.00 ± 1.01	28.00 ± 1.12
Bacteria count (Count/100)	-	4.2 x 10 ²	3.1 x 10 ²

ESTDEV – Standard Deviation

Table II: Levels of Heavy Metals in Seawater

Parameters Heavy metals/Units	WHO Value (EST DEV)	Wet Season (Mean Value) ± ESTDEV	Dry Season (Mean Value) ± ESTDEV
Na (mg/L)	-	1359.00 ± 2.13	1359.00 ± 2.13
K (mg/L)	-	106.50 ± 0.13	106.50 ± 0.13
Ca (mg/L)	NS	326.00 ± 1.21	320 ± 1.20
Mg (mg/L)	20	51.17 ± 0.13	50.20 ± 0.14
Fe (mg/L)	1	1.51 ± 0.13	0.02 ± 0.001
Pb (mg/L)	0.01	0.013 ± 0.01	0.013 ± 0.01
Cr (mg/L)	0.05	-	-
Hg (mg/L)	0.05	-	-

ESTDEV – Standard Deviation

Temperature and salinity are the two most important properties of seawater (salinity has to do with the concentration of the dissolved salts) for they control the density warm water will tend to have higher salinity than cooler water because of evaporation (Brown, and Hammond, 1961; Malton Keynes, 1997). Both were higher during the dry season. The colour was clear during the wet season 9.20 and 11.30 during Hazen unit. The total dissolved was 26,001.00 mg/L wet season and 23,000.00 mg/L dry season. These values could be due to run-off from rivers and wave actions of the season. The electrical conductivity was high due to the presence of dissolved salts (contributed by sodium, potassium, calcium and magnesium ions). It was higher during the dry season due to evaporation. Horne (1978). These values were higher than those recorded by Courant, Poweh, and Michael, (1985). Alkalinity was between 41.00 – 43.00mg/L due to slake lime CaCO_3 and Acidity values 8.50 – 10.30mg/L. Higher values were obtained for acidity during the wet season, while alkalinity was higher during the dry season. The pH ranges between 7.6 – 7.8 showing slight alkalinity.

Nitrate value was 0.13 – 0.18 mg/L, phosphate 0.269 – 0.321mg/L. Phosphates are utilized by phytoplankton and Ammonium which is oxidised to nitrate formed during bacteria decomposition of organic matter. Total bacteria count was small due to the saline nature of the seawater. Chloride values were 2100.10 – 1299.80mg/L with higher values during the dry season, these values were expected also because of the salty nature of the sea water. Dissolved oxygen values were 4.20 – 4.70, Biological Oxygen Demand was 2.80 – 3.01 while Chemical Oxygen Demand was 821 – 837 mg/L due to pollution by non-oxygen dependent bacteria leading to toxic water condition due to oil waste. Sodium, cadmium, potassium and magnesium were determined to be 1359.00mg/L, 106.50 mg/L, 320.00-326.00 mg/L and 50.20 - 51 mg/L respectively. Iron was 0.015 – 1.51, while lead, chromium and mercury were below detectable limit. Iron was higher during the wet season because of inflow from rivers and runoffs. All the parameters determined except pH, total alkalinity and nitrate were above World Health Organisation (2003) standard limit for drinking or potable water, Standard Organisation of Nigeria (2003) and Federal Environmental Protection Agency.

Conclusion

The analysis of the water quality parameters of Ogulahai seawater in Bomadi Local Government Area of Delta State shows that some of the parameters are above the World health Organisation limit for drinking/potable water. It is therefore not a source of potable water for the Ogulahai community.

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GEOPHYSICAL INVESTIGATION OF EFFECTS OF TOPOGRAPHIC COMPLEXITIES ON GROUNDWATER POTENTIAL IN IBUSA, DELTA STATE NIGERIA

Okolie, E.C

Department of Physics
Delta State University Abraka, Nigeria
okoliepeace@yahoo.com
234-0803 678 4353

Abstract

Ibusa is situated a few kilometres from the River Niger near the State capital, Asaba. Despite its nearness to the course of River Niger, it is positioned on a cliff which makes the acquisition of groundwater a serious problem for most of the inhabitants. It marks a transition point from the river bank to the hinterland and between geological formations. It is therefore, necessary to carry out geophysical survey within and around Ogboli, Ezukwu and Achala in Ibusa to determine the topographic complexities and their effects. Thus, a sensitive SAS 1000 terrameter was used to make ten VES soundings using Schlumberger array. The results indicate that Ogboli and Ezukwu villages have typically deep aquifers which are enclaved by deep rocks while neighbouring Achala village is a low land zone with shallow aquifer. In addition parts of Ogboli consists of thick lateritic top soil to about 10 m, a remarkable weathered rocks of high iron content at shallow depths of 15 – 18 m and a thick formation of hard granite at far depth. Perched aquifer exists at shallow depths of 30 – 40 m. Viable aquifer in Ogboli and Ezukwu are at 80 – 110 m but Achala possess loose top soil to a depth of 18 – 20 m followed by lateritic soil formation of about 4.5 m thick, a layer of medium to gravely sand and shallow aquifer at about 30 – 40 m

Key words: Topographic complexities, Schlumberger, formations, aquifers, Ibusa Nigeria

1.0 INTRODUCTION

The problem of potable water for domestic and industrial utilities has generally been an age long issue in Ibusa despite its nearness to Asaba, the bank of river Niger where groundwater is easily obtained. A number of boreholes have been dug in Ibusa without success. In some cases many trial wells are drilled before seeming success is achieved in Ogboli and Eziukwu villages for example. However, this problem does not exist in Achala which is about 2 km away. This work suggests that these variations could be due to topographic changes. Hence, a geophysical study was initiated to ascertain the effects of topographic complexities and determine the depths of viable aquifers in these villages in Ibusa. The study was made in Ibusa, Delta State Nigeria using a sensitive Signal Averaging System (SAS) 1000 terrameter and the field data were analysed using qualitative and quantitative methods from which the geoelectric sections of the area were obtained

1.1 LOCATION OF STUDY AREA

Asaba is the capital of Delta state. It is situated on the western bank of the River Niger while Onitsha is on the eastern bank. A few kilometres still on the west of Asaba is Ibusa. Precisely, Ibusa is located about eight kilometres west of Asaba. It is within Longitude $6^{\circ}10'N$ and $6^{\circ}13'N$ and Latitude $6^{\circ}32'E$ and $6^{\circ}34'E$ (Fig 1). It is bounded by a number of small streams from heterogynous sources which indicate that Ibusa is on a cliff. Its nearest neighbours are Ogwashi-Uku to the west and Okpanam to the north. It is typically a geologic zone.

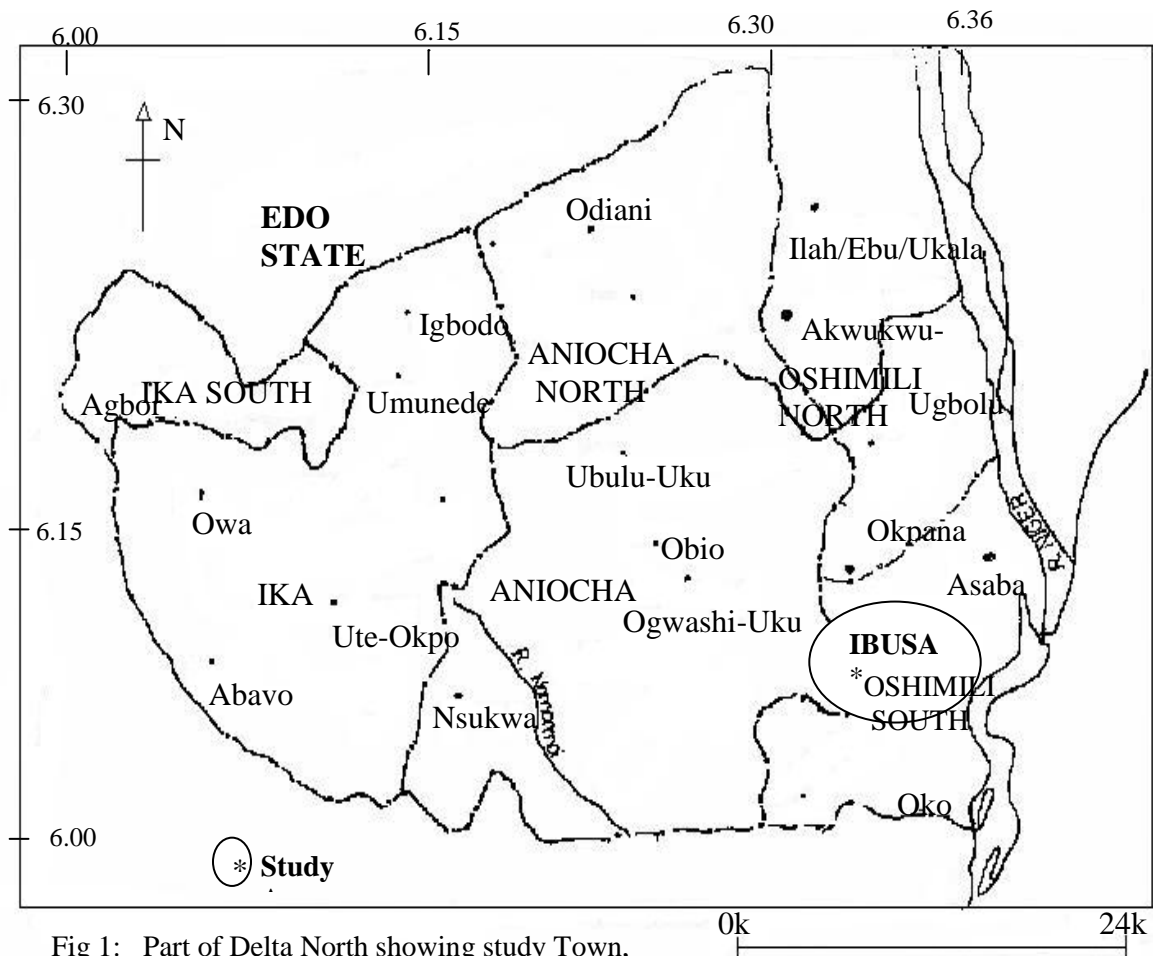


Fig 1: Part of Delta North showing study Town, Ibusa

2.0 MATERIALS AND METHOD

The vertical electrical sounding (VES) was used to determine the electrical resistivities and depths of the subsurface layers with a sensitive ABEM SAS 1000 terrameter. On the whole, ten VES stations were established and surveyed in three neighbouring villages at Ibusa using the Schlumberger array. The Schlumberger array of electrical resistivity method was applied due to its relatively low cost of field operation, logistics of reduced man power and reliability on application to formation and groundwater investigations (Ako and Osundo, 1986).

On taking a sounding, the terrameter sends current into the earth through a pair of conducting electrodes, automatically computes and displays the apparent resistivity of the subsurface structure under investigation Dobrin, (1976).

Generally, the arrangement consists of a pair of current electrodes and a pair of potential electrodes which are driven into the subsurface to make a good contact with the earth in a particular site of interest (Fig 2).

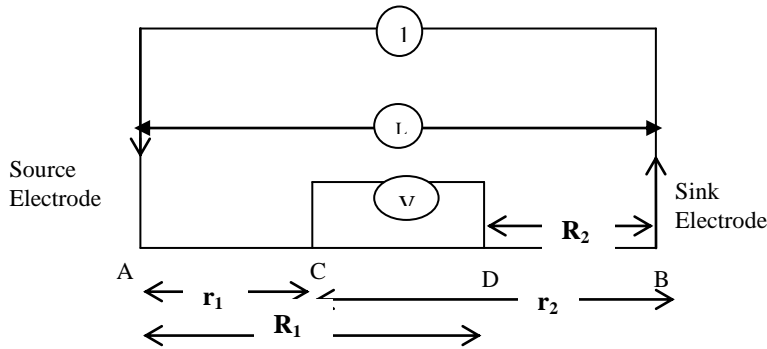


Fig 2: General four-electrode configuration for resistivity survey.

Thus, the potential difference ($V_c - V_D$) between the two inner electrodes measured by the voltmeter connected between C and D (Keary and Brooks, 1991) is

$$\nabla V = (V_c - V_D) = \frac{\rho}{2\pi} \left\{ \left(\frac{1}{r_1} - \frac{1}{r_2} \right) - \left(\frac{1}{R_1} - \frac{1}{R_2} \right) \right\} \dots\dots\dots 1$$

Hence, the subsurface resistivity by Griffith and King, (1976) is

$$\rho = 2\pi \frac{\Delta V}{I} \left\{ \frac{1}{\left(\frac{1}{r_1} - \frac{1}{r_2} \right) - \left(\frac{1}{R_1} - \frac{1}{R_2} \right)} \right\} \dots\dots\dots 2$$

$$\Rightarrow \rho = 2\pi r \left\{ \frac{1}{\left(\frac{1}{r_1} - \frac{1}{r_2} \right) - \left(\frac{1}{R_1} - \frac{1}{R_2} \right)} \right\} \dots\dots\dots 3$$

The apparent resistivity is obtained since formation measurements are not made directly (Okolie, et al 2006). Moreover the wider the electrode spacing, the deeper is the current penetration. Current penetration to a depth say Z achieved with a current electrode spread L, (Fig 2) (Okwueze and Ezeanyim, 1985) is given by

$$L = 3Z$$

$$\Rightarrow Z = L/3 \dots\dots\dots 4$$

In this work, the Schlumberger array was used to ensure deep penetration and for logistics of limited man power in the field. The Schlumberger array required that the current electrode spacings are increased on a logarithmic scale while the potential electrodes are kept at small separations relative to the current electrodes separations (Fig 3) ensuring that $AB \geq 5CD$ (Lowrie, 1997). Thus, only current electrodes need to be shifted to new position for most readings while potentials electrodes are kept undisturbed for up to three or four readings. The current and potential pairs of electrodes therefore have a common midpoint O, but the distance between adjacent electrodes differs (Oseji, et al 2005).

Hence, the potential at electrode P_1 from C_1 (Fig 3) (Zohdy, 1988).will be

$$V_{P_1} = \frac{\rho I}{2\pi} \left\{ \frac{1}{a - b/2} - \frac{1}{a + b/2} \right\} \dots\dots\dots 5$$

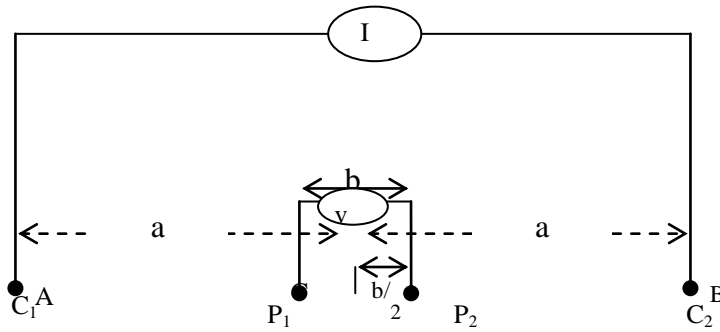


Fig 3: Schlumberger field electrode arrangement

where “a” is the distance between the current electrode and station midpoint, “b” is the distance between potential electrodes and “2a” is the current electrode separation.

And the potential at P₂ from C₁ is

$$V_{P_2} = \frac{\rho I}{2\pi} \left\{ \frac{1}{a + b/2} - \frac{1}{a - b/2} \right\} \dots\dots\dots 6$$

where $a \gg b$ as in Schlumberger array.

The potential difference “dV” between the two potential electrodes is therefore,

$$dV = \frac{\rho I}{2\pi} \left(\frac{8b}{4a^2 - b^2} \right) \text{ becomes}$$

$$= \frac{\rho I b}{\pi a^2}$$

$$\text{and } \rho_{as} = \frac{\pi a^2}{b} \frac{dV}{I} = \frac{\pi a^2}{b} R \dots\dots\dots 7$$

where ρ is apparent resistivity for Schlumberger array and Geometric factor for Schlumberger array is $K_s = \frac{2\pi}{8b} (4a^2 - b^2)$ (Okolie, *et al* 2008).

$$\text{Hence, for } a \gg b, K_s = \frac{\pi a^2}{b} \dots\dots\dots 8$$

The apparent resistivity values (Table 1) recorded by the field tarrameter were plotted against half current electrode spacing on a 3- decade bi-log graph from which the qualitative and quantitative analyses were made using partial curve matching technique to obtain the apparent resistivity replacement and depth Index of each formation in the sites. These were matched with corresponding master and auxiliary curves and the results were used to perform and obtain Resist software computer iteration for effective analysis and formation stratification and interpretation (Fig 5 – 9) (Okwueze, *et al* 1988).

Table1: Sample Field Data (Apparent Resistivities in Study Area, Ibusa)

MN/2 (m)	AB/2 (m)	Eziukwu VES 1 (Ω m)	Ogboli VES 1 (Ω m)	Ogboli VES 2 (Ω m)	Achala VES 1 (Ω m)	Achala VES 2 (Ω m)
0.2	1.00	78	118	150	75	62
	1.47	85	127	162	81	84
	2.15	97	178	130	76	108
	3.16	132	240	148	95	124
2/1.0	4.64	216	148	218	115	147
	6.81	105	128	242	128	110
1.0/3.0	10.00	168	172	294	189	128
	14.70	207	235	321	203	157
	21.50	239	398	426	130	132
	31.60	344	710	585	296	146
3.0/8.0	46.4	324	761	843	368	215
	68.10	327	1356	895	347	235
8/16	100.00	398	2563	1256	296	255
	147.00	538	1987	2272	521	219
16/30	215.00	853	1845	3532	772	405
30/50	316.00	715	3431	2188	943	278
	464.00	947	5613	1095	650	195

3.0 RESULTS AND DISCUSSION

The results show that virtually all sites in the study area exhibit A – type curve (Fig 5 – 9). However, while Ogboli and Eziukwu consist of nine distinct formation strata, Achala has seven layers. Also, Ogboli and Eziukwu possess loose top soil of 3m, a layer of thick lateritic soil to about 10 m, shale, remarkable weathered rock of high iron content at shallow depths of 15 – 18 m and thick rock shield at far depth of about 70 - 80m. Perched aquifer with high iron content exists at shallow depths of 30 – 40 m in Ogboli and Eziukwu, while viable aquifer is within 80 – 110 m and typically enclaved by deep rooted rock shield. In contrast, neighbouring Achala is a low land zone has fine top soil, a thin layer of lateritic soil clayey sand, silt and perched aquifer at shallow depths of about 10 metres, a layer of medium to gravely sand and

shallow aquifer at about 30 – 40 m. Its main aquifer is at about 30 metres (Figs10&11) Okolie *et al* (2007).This is in consonance with monitored direct log data.

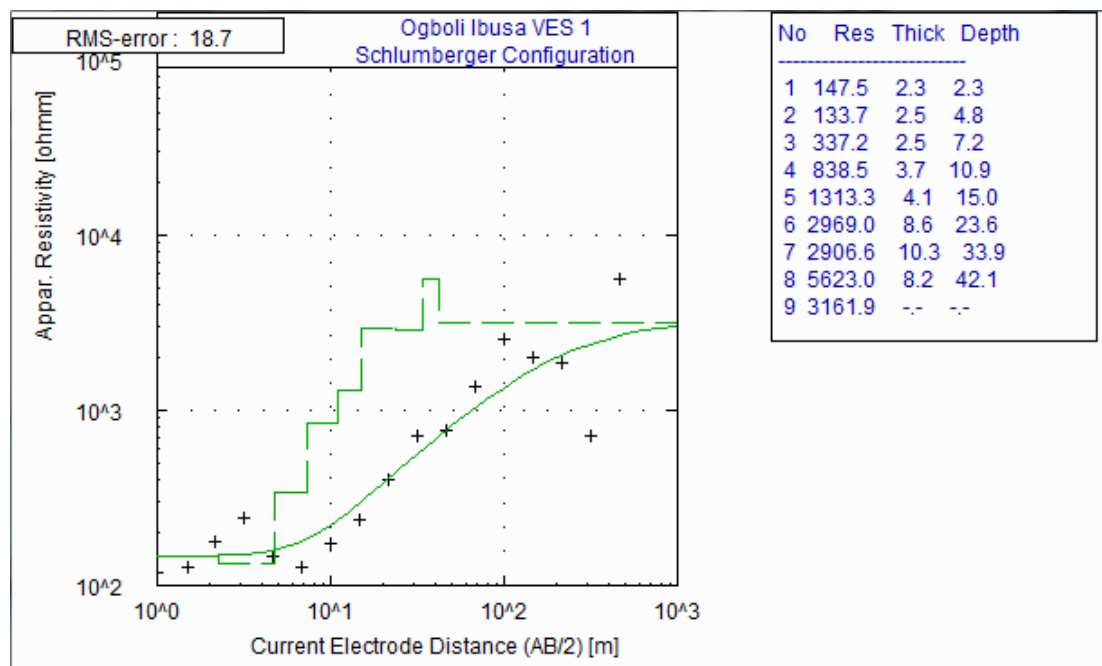


Fig 5 (colour online): Sample Plot for Site 1 in Ogboli, Ibusa

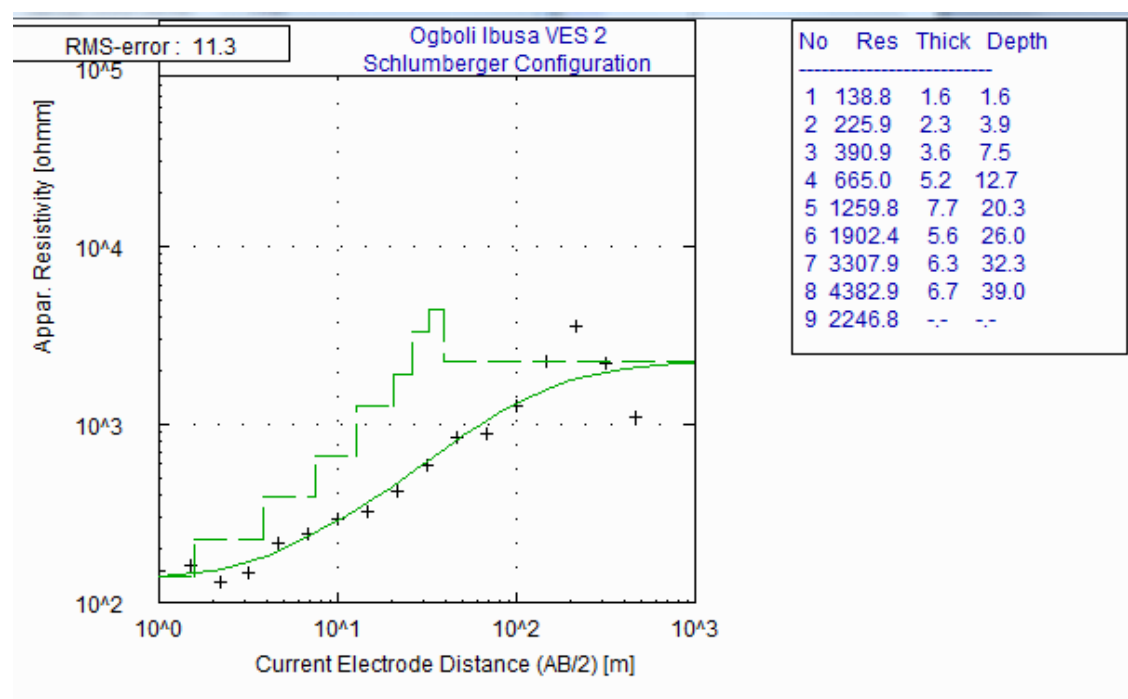


Fig 6 (colour online): Sample Plot for Site 2 in Ogboli, Ibusa

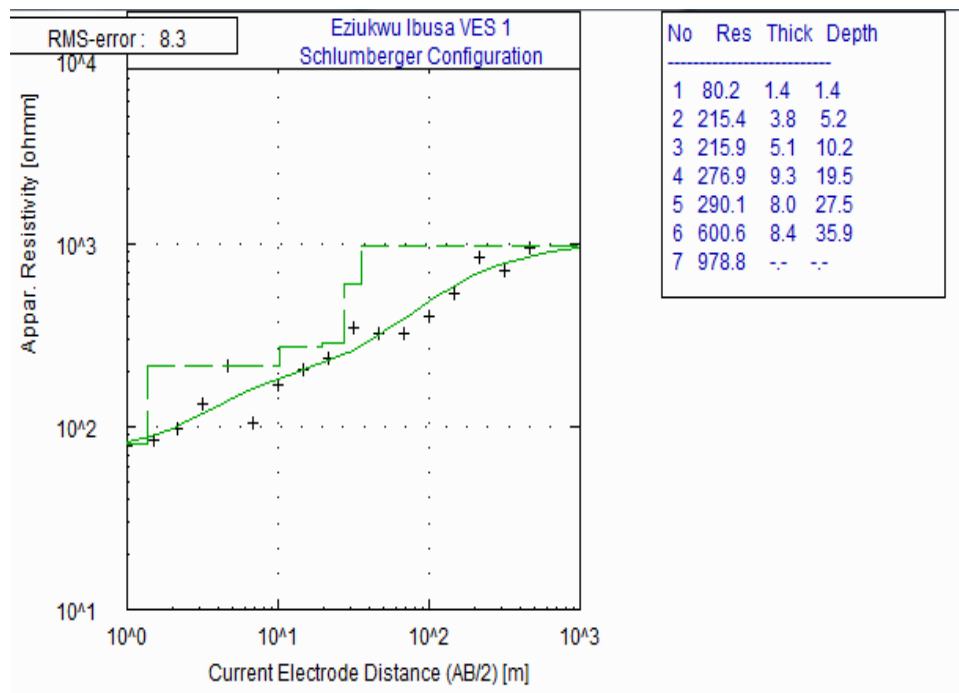


Fig 7 (colour online): Sample Plot for Site 1 in Eziukwu, Ibusa

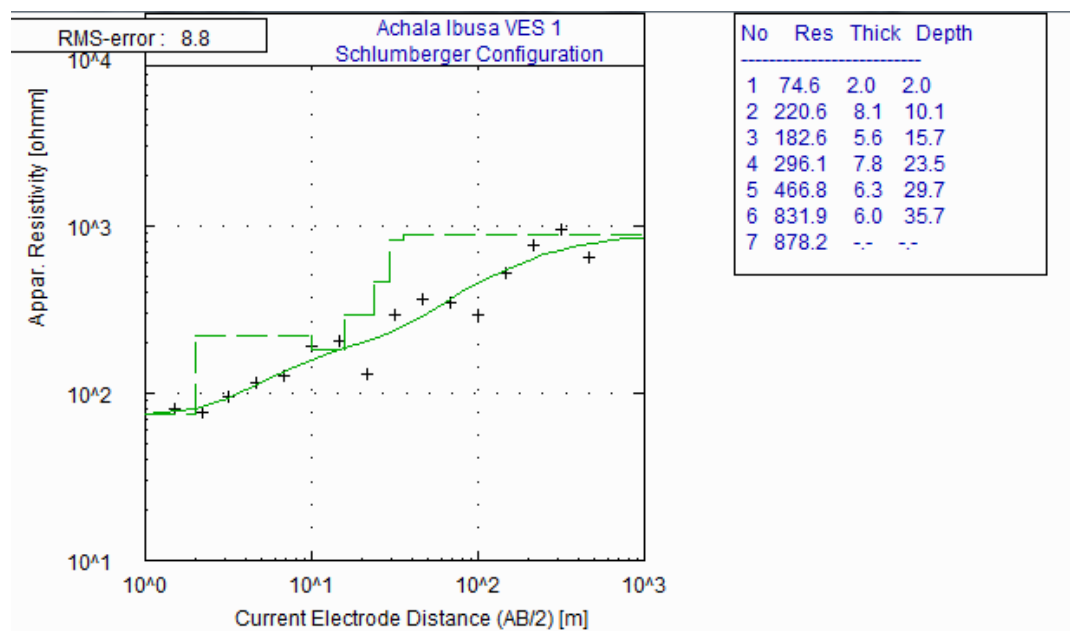


Fig 8 (colour online): Sample Plot for Site 1 in Achala, Ibusa

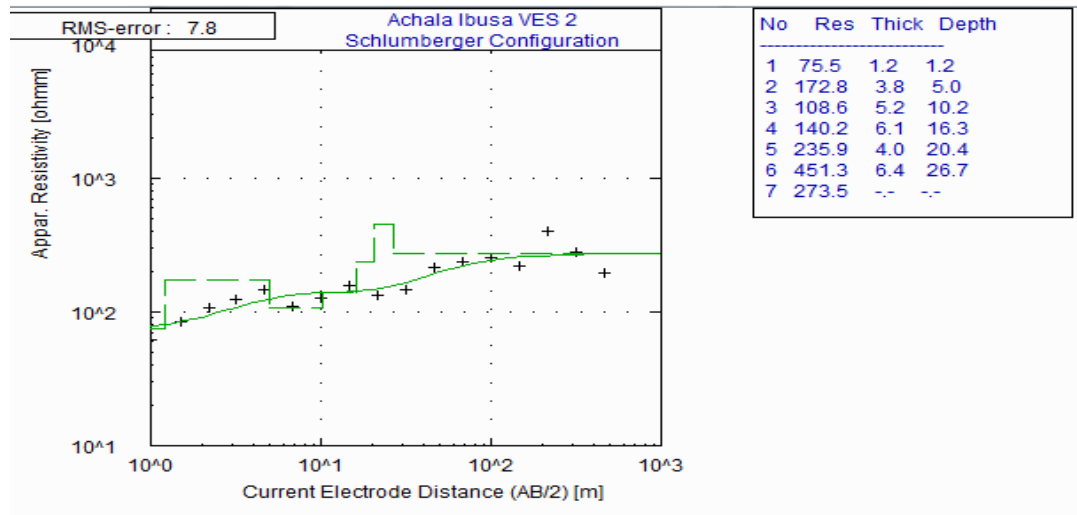


Fig 9 (colour online): Sample Plot for Site 2 in Achala, Ibusa

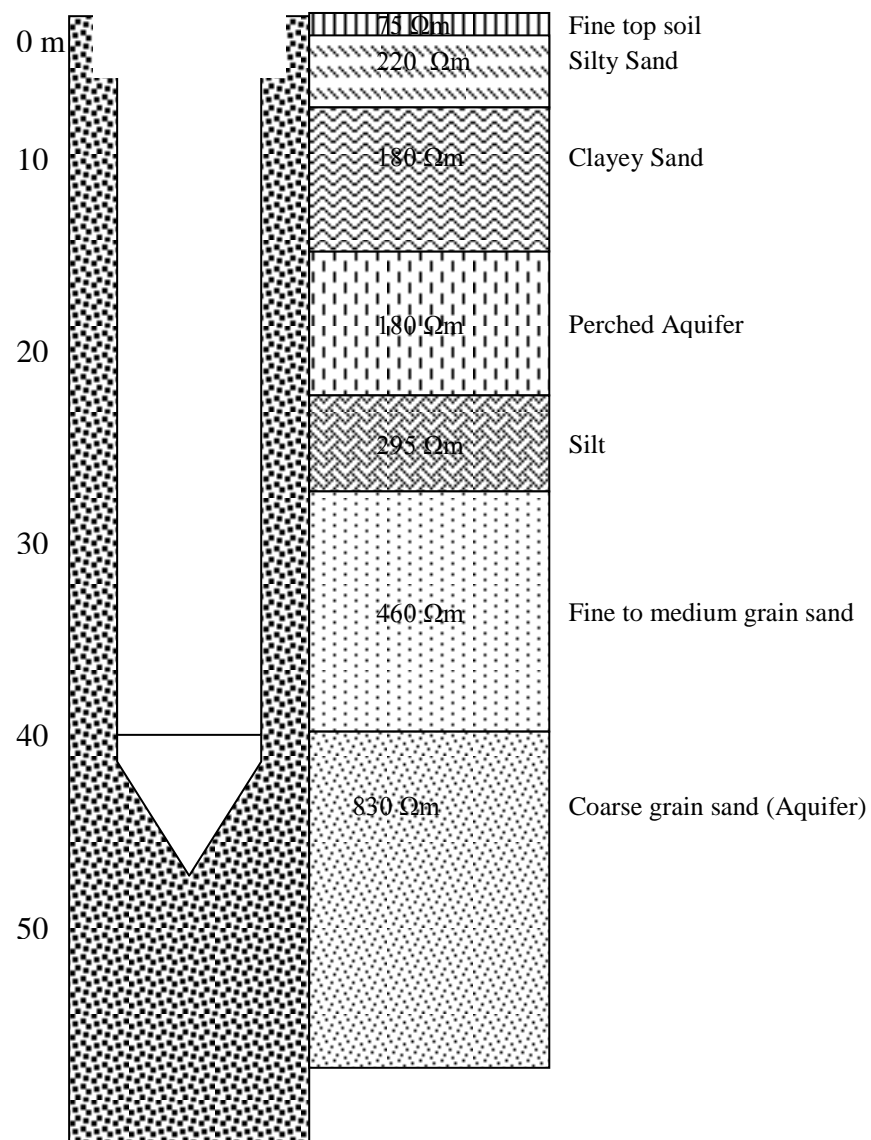


Fig 10: Geoelectric section of Achala in Ibusa

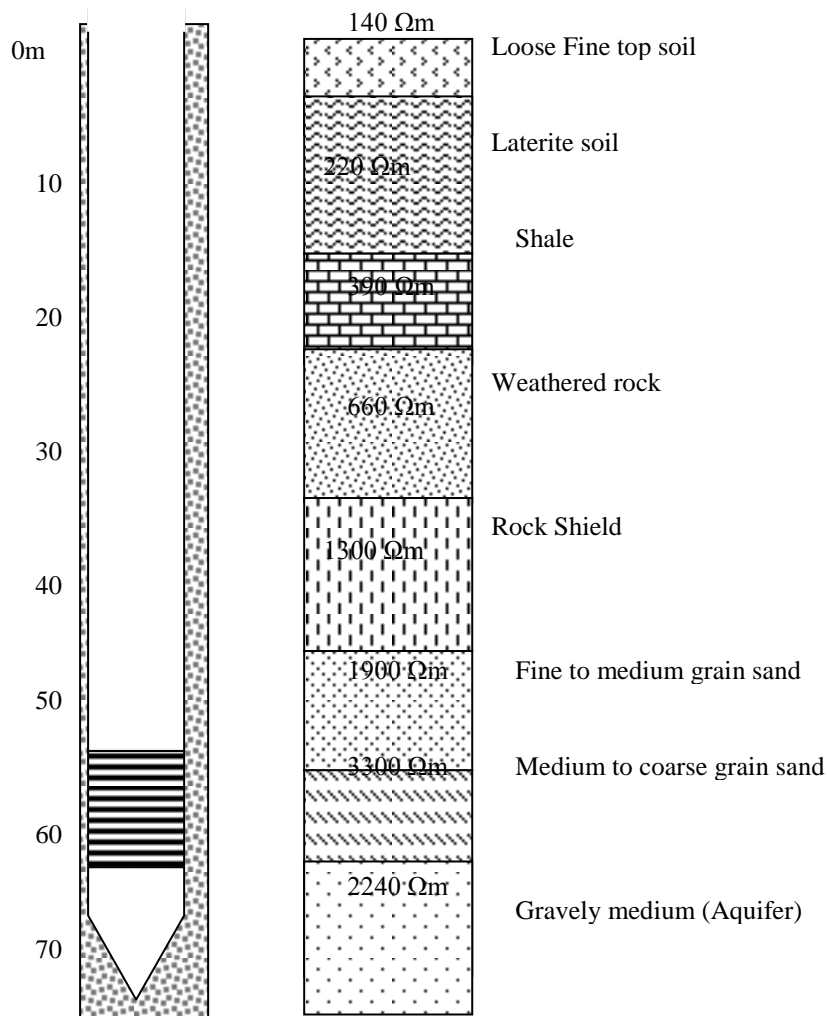


Fig 11: Geoelectric Section of Ogboli & Eziukwu, Ibusa

4.0 CONCLUSION AND RECOMMENDATION

Subsurface formation at Ibusa is a blend of basement complex and sedimentary terrain. Virtually, all sites in Ogboli, Eziukwu and Achala exhibit A-curve which indicates that the curve type here is invariant with respect to topography.

The geoelectric section (Fig 10) shows promising aquifers are readily available in Achala where low resistivity subsurface strata due to the presence of clayey sand and its relative closeness to River Niger. These sites at Achala are indicative of high water bearing medium (aquifer) with medium to coarse grain sand at 25 – 30 m depth. Perch aquifers also exit at shallow points of about 12 m. Sites around Ogboli and Eziukwu consist of high resistive formations to far depth with no distinct aquifer. These sites also possess rock shield formation at far depth which may be associated to the sudden change in topography as one drifts away from the bank of river Niger (Fig 11). It is therefore recommended that for effective bore hole siting in the study area sites on the East end of Ogboli and Eziukwu as well as those in Achala should be the target. This will ensure long term continuous supply for both domestic and industrial utilities for many a people living in Ibusa and within Asaba metropolis.

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APPLICATION OF 3-D STRUCTURAL INTERPRETATION TO RESERVOIR DESCRIPTION: FIELD 'X' EASTERN NIGER DELTA.

G.U. Ozulu¹ and M.O. Ofomola²

¹Department of Physical Sciences, Novena University, Ogume, Delta State, Nigeria.
georgedquest@yahoo.com

²Department of Physics, Delta State University, Abraka, Delta State, Nigeria.
ovirimerrious@yahoo.com

Abstract

An attempt has been made to use structural interpretation to describe the reservoirs of field 'X' eastern Niger Delta using an integration of well log and 3-D seismic data. Electric log method was used to structurally correlate three wells: 'X' _ 01, 04 and 05 and this helped to mark the base of the fresh water sand (BFS) at a depth of – 7920ft (MD). Four reservoir sand units (CO₁, DO₁, DO₆ and DO₇) within the Agbada Formation were penetrated by the three straight hole wells up to a total depth of 13,000 feet (MD) in well 'X' _ 01. Petrophysical analysis shows porosity values range from 19 – 29%, permeability from 379 – 613md, and transmissivity from 13,644 – 77,238. Two reflecting horizons picked during seismic interpretation were tied to the CO₁ and DO₇ reservoirs at time 2476ms and 2780ms respectively and were then mapped for the structural analysis. Depth structural maps prepared for the top of CO₁ and DO₇ sand units defined the structural architecture of the reservoirs as plunging domal anticlines with a 3-way fault sealing structural closures trapped against the NW-SE antithetic fault. Reservoir volumetrics show the rock volume for the CO₁ reservoir sand to be 201,122.67 acre – ft while the recoverable oil reserve stands at 85, 624, 993.95 bbl.

Key words Well-log. Correlation. Seismic. Structural. Reservoir. Field.

Introduction

Geologic reservoir description is the act of putting all relevant geologic data in order to give a good visualization of the geologic domain of the reservoir. To achieve this, a map version of the reservoir may be constructed, a grid-block version or 3-D visualization of the reservoir produced.

Jardine and Wilshart (1987), stated that in order to produce good model of reservoirs, continual revision of the data is required as depletion of the reservoir proceeds and more data become available.

Haldorsen and Damsleth (1993) stated that seismic interpretation aid in reservoir architecture mapping. According to them, seismic data do not only find fields they also reveal the lateral and vertical extent. The interpretation of these data provides the reservoir surface topography (i.e. depth map for top reservoir and sometimes for different horizons within it). It also helps in locating the structural and the stratigraphic traps as well as the structural closures and spill points.

Corvey and Cobley (1992), showed how reservoir descriptions are developed. According to them, the first stage is that of defining the reservoir's large scale structures using deterministic data. This stage aims at constructing as detailed a geologic description of the reservoir as measurement will allow. One of such vital information needed, is the structural interpretation from seismic data.

As a result of the confidential nature and the high risk involved in disclosing information in the oil and gas industry, the actual names of the location wells and the field of the study area has been concealed. The field, however, has been designated as Field 'X' Eastern Niger Delta, while the location wells are named wells 01, 04, and 05. The study area is located on the north-eastern onshore part of the Niger Delta Petroleum Province Nigeria which is a part of Nigeria's oil concessions.

The Niger Delta is situated on the continental margin of the Gulf of Guinea in equatorial West Africa, between latitude 3° and 6° N and longitude 5° and 8° E. It extends throughout the Niger Delta province and ranks amongst the world's most prolific petroleum producing Tertiary deltas that account for about 5% of the world's oil and gas reserves and for about 2.5% of the present day basin area on earth (Reijers et.al, 1997).

The Geology of the Niger Delta shows that the onshore portion is delineated by the Geology of southern Nigeria and Cameroon. The northern boundary is the Benin flank, an east-northeast trending hinge line south of the West African basement massif. The northeastern boundary is defined by outcrops of the Cretaceous on the Abakaliki High and further east to the southeast by the Calabar flank, a hinge line bordering the adjacent PreCambrian. (Fig.1)

The oil producing Niger Delta region is underlain by Quaternary to Recent sedimentary formations (Short & Stauble, 1967; Weber & Daukoro, 1975).

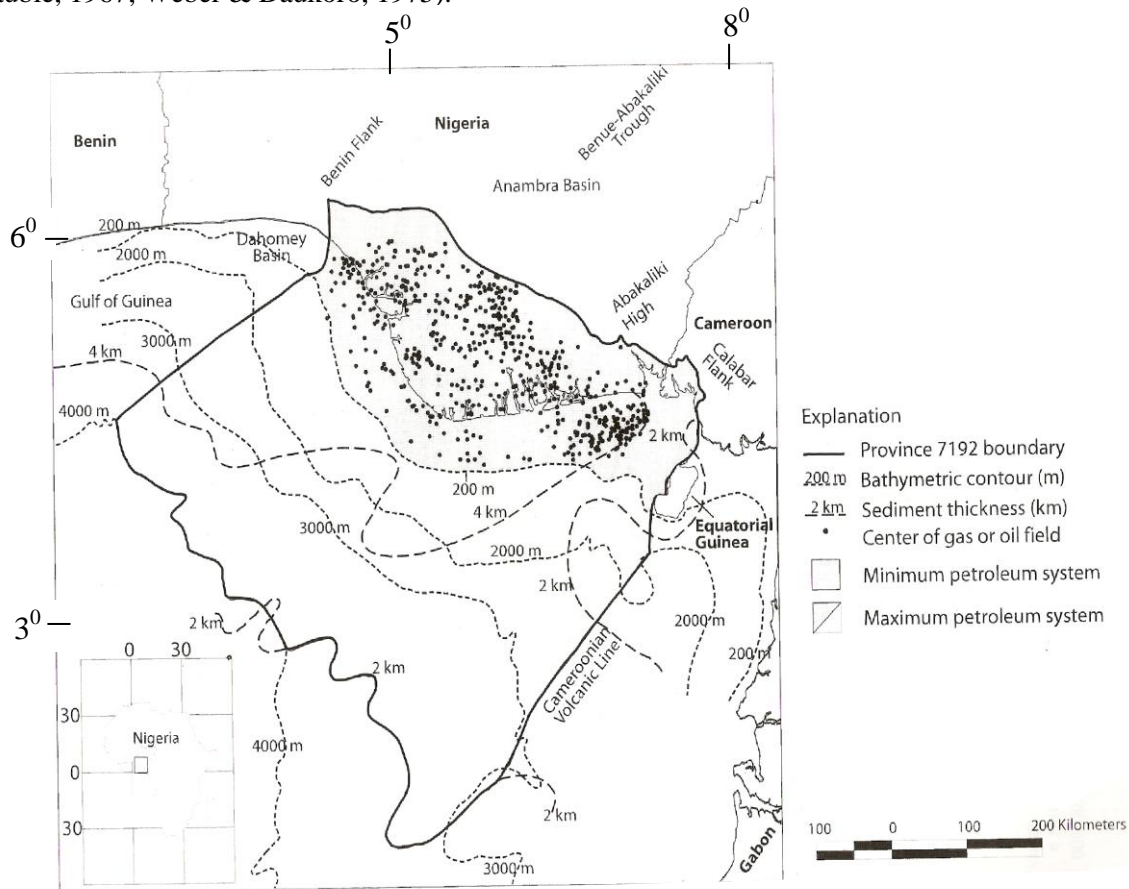


Fig. 1 Index map of Nigeria and Cameroon showing province outline

Three main lithostratigraphic units have been recognized in the Tertiary Niger Delta. These represent prograding depositional facies distinguished mostly on the basis of their sand-shale ratio. According to Short and Stauble (1967), well sections generally display these vertical subdivisions. These are the pro-delta marine Akata Formation (base), the paralic Agbada Formation (middle) and the continental Benin Formation (top).

According to Selley (1998), if seismic surveys are shot on grids of say 50m or so, then a 3-D matrix data is acquired that enables seismic displays to be produced not only along the survey lines shot, but also in any other orientation. This is called 3-D seismic. Horizontal displays or time slices as well as seismic cubes can be produced from the data.

This paper presents a description of the main reservoirs in Field 'X' utilizing well-log structural correlation, petrophysical analysis as well as well-to-seismic matching to interpret the reservoir structures.

Methodology

Wire-line well logs (composite logs) from three wells (01, 04 and 05) drilled in the field were used for the structural correlation using the stratwork software. These include the gamma-ray (GR) log used for lithologic correlation, the long normal and short normal (LLD and LLSD) resistivity logs used for the determination of the hydrocarbon bearing zones. The neutron-porosity (NPHL) and formation density (RHOB) logs were used to determine the type of hydrocarbons (oil or gas).

Seismic fault interpretation and horizon interpretation were done using the seiworks. The faults were interpreted on the dip lines and confirmed on the traces. Spectrum colour bars were used as indicators to pick strong reflection horizons.

The structural and stratigraphic correlation was done for three wells that penetrated the Benin and the Agbada Formations. The first major thick shale lithologic unit was used to define the boundary between the Benin and the Agbada Formations. The top of the shale corresponds to the base of the massive sand from the Benin Formation and was annotated as the base of the fresh water sand (BFSW). This boundary was marked at a depth of -7,920 ft (MD).

Seven sand units referred to as sand facies or sand lithologies were identified from the log analysis. These were annotated as A, BO₁, BO₇, CO₁, DO₁, DO₆ and DO₇. All these are found in the Agbada Formation. Out of these reservoir sands, CO₁, DO₁, DO₆ and DO₇ were quite distinct and penetrated up to a depth of 13,000 ft (MD).

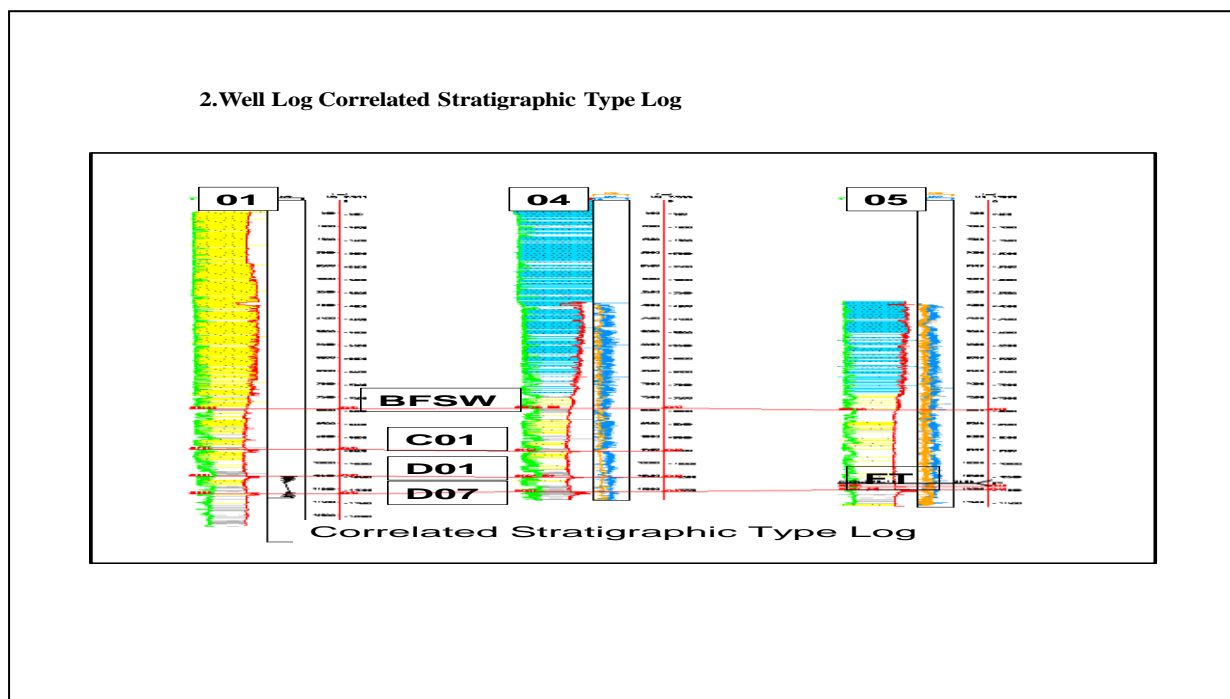


Fig 2 (colour online): correlated stratigraphic type log for field 'X'

The syntool was used to produce a seismic backdrop in order to carry out well-to-seismic matching (tying well data to seismic). This aided in bringing seismic lines and superimposing them on well logs on the 3-D gridded seismic base map through the process of digitization. After that, all relevant information like fault traces and horizons were imported from seisworks to syntool. A zone control map was then created. This comprised the Master File Directory (MFD) housing all information. A Control Gridded File (CGF), which is a picture file, was also created. All these helped in generating the fault polygon using the Z-Map plus. In order to construct the depth structural maps of the field, the field velocity model was built using the three correlated wells and their check-shot. This helped to convert the data from time to depth using the Time-Depth conversion model (TDQ) software. The horizon data points were then exported along with the fault

polygon back to the Z-Map plus application software for final contouring and generation of the depth structure maps.

Structure contour line, net isopach line for the top of CO_1 reservoir sand and square counter values generated using the square counter grid method were all used to obtain the area circumscribing the top of the main reservoir sand (CO_1). The Trapezoidal rule was then applied to determine the reservoir volumetric.

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Table 1. Petrophysical Data Sheet for Well 'X'- 04 WELL: 'X'-04 V SHALE : >0%<40% LOCATION KB : 42' POROSITY: >12%<40% X=00 WELL TYPE: STRAIGHTHOLE Sw : >0%<70% Y=00												
RESV. SAND	TOP OF SAND (MD)	BASE OF SAND (MD)	GROSS SAND	NET SAND	NET PAY	FLUID CONTACTS	AVG. POROSITY (Φ)	PERMEABILITY (K) md	TRANS MISSIVITY Y (Kh)	AVG. H ₂ O SAT. (Sw)	AVG. HC SAT. (Sh)	REMARK
CO ₁	9558	9594	36	32	27	OWC@9618	0.29 (29%)	379	13644	0.18 (18%)	0.82 (82%)	OIL
DO ₂	10540	10630	90	74	32	GOC@10570 OWC@10597	0.26 (26%)	532	47880	0.31 (31%)	0.69 (69%)	GAS & OIL
DO ₇	11118	11244	126	88	62	OWC@11264	0.25 (25%)	613	77238	0.22 (22%)	0.78 (78%)	OIL
121 FT OF OIL IN THE THREE RESERVOIR SANDS												
Table 2. Petrophysical Data Sheet for Well 'X'- 05 WELL: 'X'-05 V SHALE : >0%<40% LOCATION KB : 45' POROSITY: >12%<40% X=00												
RESV. SAND	TOP OF SAND (MD)	BASE OF SAND (MD)	GROSS SAND	NET SAND	NET PAY	FLUID CONTACTS	AVG. POROSITY (Φ)	PERMEABILITY (K) md	TRANS MISSIVITY Y (Kh)	AVG. H ₂ O SAT. (Sw)	AVG. HC SAT. (Sh)	REMARK
DO ₆	10896	10934	38	32	30	LKO@10934	0.19 (19%)	388	14744	0.29 (29%)	0.71 (71%)	OIL
DO ₇	11010	11112	102	90	87	LKO@11112	0.24 (24%)	613	62526	0.15 (15%)	0.85 (85%)	GAS & OIL
117 FT OF OIL IN THE TWO RESERVOIR SANDS												

3 Results and Discussion

The results of the petrophysical analysis are shown in tables I and II. Porosity values range from 19-29%, permeability range from 379-613 md and transmissivity from 13,644-77,238.

Figure 3 clearly shows the structural style of the field. The major structure bounding the field is the normal fault (green fault) trending NW-SE. The minor faults which occur are both antithetic and synthetic to the major faults. These growth faults are typical of fields in the Niger Delta. The synthetic fault (pink fault) and the major structure building fault (green fault), show a bifurcating fault pattern while the antithetic fault (blue fault) and the major structure building fault, form a compensating fault pattern.

Matching seismic into the well log data, two designated formation top of the CO₁ reservoir corresponded to horizon H₁ and the DO₇ reservoir corresponded to horizon H₂. The yellow picked horizon was the CO₁ with a time lap of 2476ms while the green picked reflecting horizon was the DO₇ with a time lap of 2780ms. See figure 3.

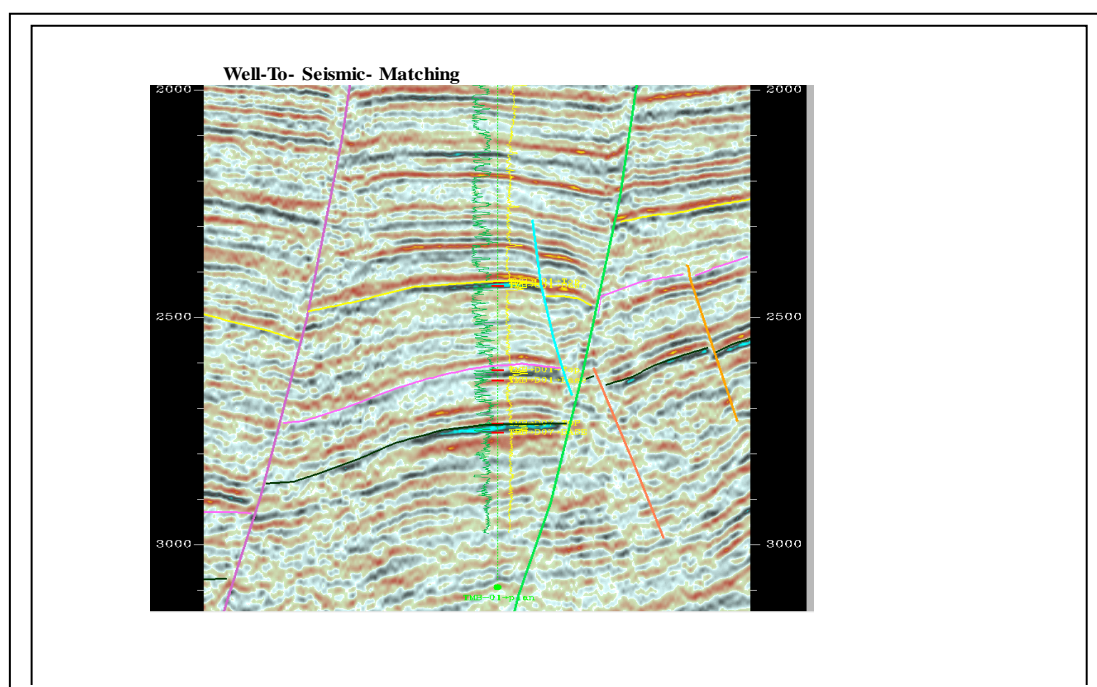


Fig 3. (colour online): Seismic dip line section showing displaced well log curves (gamma ray and resistivity), mapped horizons and interpreted faults and tops of field 'X'

Figure 4 are the depth structural maps for the top of CO₁ and DO₇ reservoir sands. The prominent structural features in the field are large plunging anticlines having a NW-SE structural trending orientation. These anticlines form the structural traps with their closures truncated by a sealing normal fault and a spill point at the eastern fringe. This is a 3-way structural closure completed by a fault as revealed by the close contour intervals in the depth structure maps.

The DO₇ reservoir has the thickest gross sand due to the occurrence of the fault and the concentration of maximum sediment deposition of the paralic Agbada Formation on the down-thrown fault block. It also has the highest capacity for reservoir performance considering the value of the transmissivity.

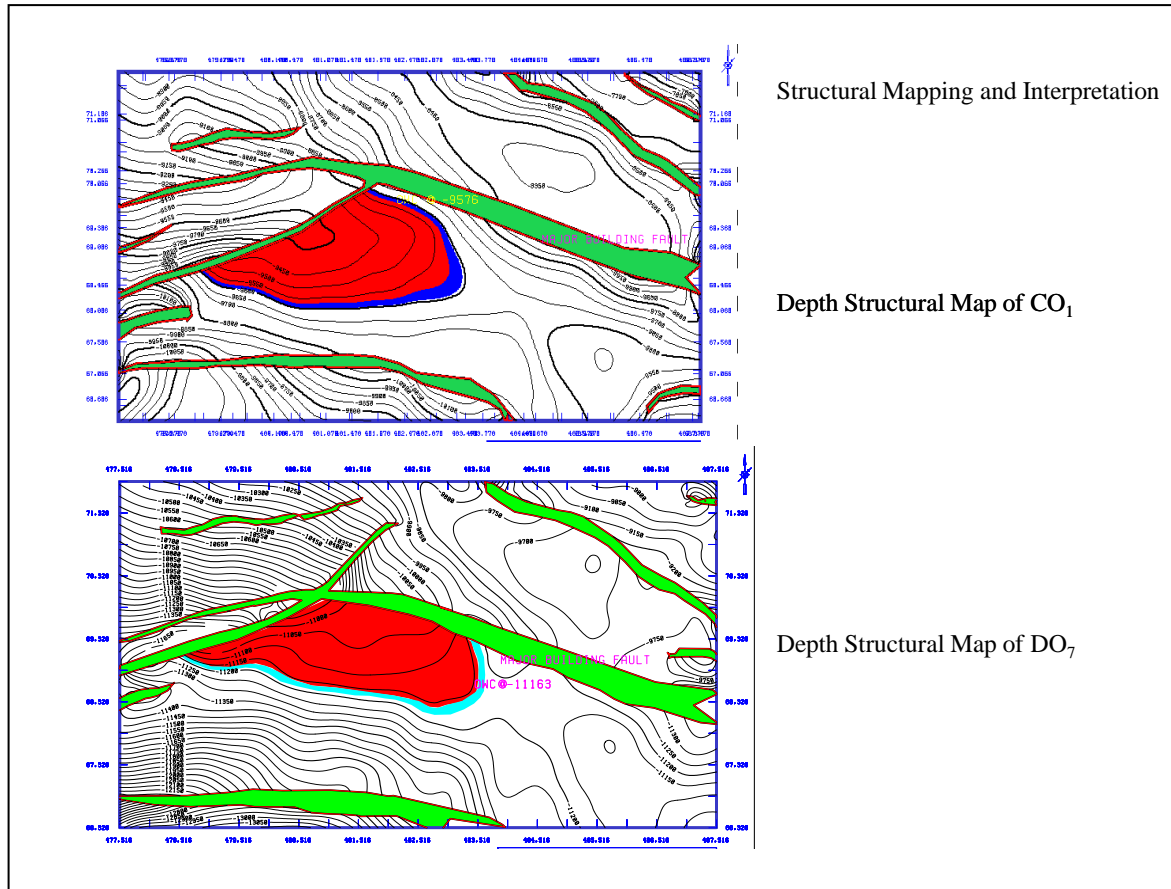


Fig 4. (colour online): Depth structural maps of CO₁ & DO₇ reservoir sands

The result of the square counter values and area of contours circumscribing the top of CO₁ reservoir was quite distinct and was used as a diagnostic reservoir to typify the volumetric estimate of the field. The Trapezoidal Rule was applied for the volumetric calculation. The formula is written as:

$$BV = (h/2)[A_0 + 2A_1 + 2A_2 + \dots + 2A_{n-1} + A_n] + h_n A_n / 2$$

Where BV= rock volume or bulk volume typically in acre-feet; h=contour interval; A₀=area enclosed by zero contour line; A₁= area enclosed by first contour line above zero; A_{n-1}= area enclosed by contour line below top contour line; A_n= area enclosed by top contour line and h_n= vertical distance from top contour to the top of the reservoir.

$$BV = (50/2)[247.744 + 2*1,107.106 + 2*1,118.719 + 2*985.17 + 189.679] + 12.5*189.679/2$$

$$BV = 201,122.67 \text{ acre-ft.}$$

The recoverable oil reserve for the CO₁ reservoir sand in (bbl) was determined by an approximation method.

$$\text{Recoverable oil (bbl)} = 7758 * V * \Phi S_{hc} * R / FVF_{oil}$$

Where V=rock volume; 7758= conversion factor; Φ =porosity (average) as a decimal from well logs or cores. S_{hc}= hydrocarbon saturation from well logs and the Archie's equation. R= recovery factor (estimated) 30 % (0.3) for sand and FVF_{oil}= formation volume factor for oil in range of 1.1 to 1.5 (an average of 1.3).

$$\text{Recoverable oil (bbl)} = 7758 * 201122.67 * 0.29 * 0.82 * 0.3 / 1.3$$

$$\text{Recoverable oil} = 85,624,993.95 \text{ bbl.}$$

Conclusion

The results have indicates that the application of 3-D Structural interpretation to the description of reservoirs in field 'X' is a modest way of visualizing the geologic domain of these reservoirs. The integration of well log and seismic data, though profitable, is still highly limited because it only gives the structural model. Static well data from core analysis, capillary pressure, production and engineering well test data could further improve the visualization of these reservoirs. Volumetric analysis could be enhanced when very useful local information on the formation volume factor (FVF), engineering well test data, among others is readily available.

In all, field 'X' eastern Niger Delta has good potential reservoirs and could form a good reserve for petroleum exploitation.

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SPATIAL DISTRIBUTION OF CLAY MINERALS IN DEEP HORIZONS WITHIN THE NIGER DELTA PETROLEUM PROVINCE

Adaikpoh E. O.

DEPARTMENT OF GEOLOGY
DELTA STATE UNIVERSITY ABRAKA
PMB 1. ABRAKA, NIGERIA.

Abstract

Clay sediments from three wells were studied using Philips X-ray diffractometer (PW-1710 Basis) and Philips XRD programme. Clay mineral contents are kaolinite, smectite, illite and chlorite in descending quantitative order. The pattern of spatial distribution of the clay minerals shows classification into three different zones representing near-shore influence in the lowermost portions (Z_1 and Z_0), deeper offshore (V – Y) basinal influence in the middle part and again near-shore/continental influence in the topmost zones (S_2 – U). The intermittent absence of illite in lithologic units (S_1 – U) suggests warm periods/events possibly due to periodic exposure. The intermittent absence of chlorite in each lithologic unit is indicative of warm period/events and also suggestive of intense chemical weathering. This study captured high occurrence of smectite which is anomalous for such depths.

Keywords: Niger Delta, Clay sediments, Anomalous smectite occurrence.

INTRODUCTION

The Niger Delta plays host to hydrocarbons on which Nigeria's economy hinges. The development of the Niger Delta and the description of its petroleum systems are documented in Allen, 1965; Burke et al, 1971; Weber, 1971; Murat, 1972; Oomkens, 1974; Weber and Daukoru, 1976; Evamy et al., 1978; Ejedawe, 1981; Whiteman, 1982; Bustin, 1988; Doust, 1989; Braide, 1993; Wu and Bally, 2000 and Corredor et al, 2005.

Clays make up about 25 – 35% of the terrigenous fraction of sedimentary rocks. During diagenesis, clay / clay minerals are precipitated out of the fluid phase and act as cementing materials that fill the pore spaces between the individual grains of the sediment undergoing diagenesis. Grim (1968) stated that kaolinite clays on reaching marine environment were altered to illite and chlorite and that montmorillonite was converted to chlorite, hence he concluded that clay minerals are environment sensitive and can be used to ascertain the nature of an ancient environment.

During clay compaction, the mineralogy changes with increasing depth. Powers (1967) produced evidence to show how montmorillonite changed to illite releasing water and generating high pore pressure while primary porosity is lost. Chlorites are also common constituents of argillaceous sedimentary rocks where these minerals occur in both detrital and authigenic forms. Where there is a predominance of chlorite implies absence of warm period and/ or nearness /or occurrence of metamorphic or igneous rocks.

Illites, which are the dominant clay minerals in argillaceous rocks, form by the weathering of silicates (primarily feldspar), through the alteration of other clay minerals, and during the degradation of muscovite (Deer and Howie, 1963). Formation of illite is generally favored by alkaline conditions and by high concentrations of Al and K, while the formation of kaolinite is favoured by acidic conditions. The presence of illite in sandstone indicates dominance of immature sandstone and nearness to source while the predominance of kaolinite group in sediments indicates the dominance of terrestrial influence (Biscaye, 1965).

Montmorillonite is a members of the smectite group which commonly result from the weathering of basic rocks. Smectite formation is favored by level to gently sloping terrains that are poorly drained, mildly alkaline (such as in marine environments).

Other factors that favor the formation of smectites include the availability of Ca and the paucity of K (Deer and Howie, 1963). Poor drainage is necessary because water can leach away ions (e.g. Mg) freed in the alteration reactions.

Pastouret et al (1978) claimed that smectite, in the Niger Delta, has its origin from the River Niger, hence an increase in marine sediments implies dominance of delta facies as a result of the reduction in the volume of discharge of the Niger River.

This study examined the distribution of clay minerals from three borehole in Umutu oil field.

AREA OF STUDY

Umutu Field is one of the oil concessions within the Niger Delta. It lies between latitude $5^{\circ}35'$ and $5^{\circ}40'$; and longitude $6^{\circ}10'$ and $6^{\circ}20'$ (Figure 1). The Umutu Field includes Umutu and Environs situated in the northwestern Niger Delta.

METHODOLOGY

Ditch cuttings from three boreholes drilled in Umutu Oil Field were first subjected to grain size analysis by sieving. Air-dried samples of less than 0.1 μ m size of the ditch cuttings were then subjected to X-ray diffraction analysis.

Pressed-tablets of the samples powder were prepared and measurements were subjected to the following equipment and configuration of instrument: CU K α -pipe, automated aperture, 3.0 {kV} voltage, 20 {mA} amperage, $\alpha_1 = 1.54060$ - $\alpha_2 = 1.54439$ {Å} wavelength differential, 2.010° start angle and 69.990° end angle, 1° /mm goniometer velocity, 2×10 amplitude, decay of 4.

One set of the samples were diffracted, then heated for 1 hour at 550°C and diffracted again while a duplicate set was solvated with glycol before diffracting. X-ray diffractograms as well as print-outs (Traces v3.0 Peak search) giving 2-theta angles, peak (counts), D-space and relative intensities were produced using Philips X-ray diffractometer (PW-1710 Basis) and Philips XRD programme (Philips PW1877 Automated Powder Diffraction, version 3.5b).

In the interpretation of the qualitative determination of mineral contents, the ASTM (Joint Committee of Powder Diffraction Standards, 1974) was used.

RESULTS

In Tables 1, 2 and 3 the relative abundance of the different clay minerals present in the boreholes are expressed as percentage of the bulk clay minerals composition. The Tables clearly show the dominance of kaolinite, smectite, illite and chlorite in descending order. These clay mineral compositions are represented in Figure 2. The figure shows generally that kaolinite has higher proportions in the sandy horizons (S_1 -U). Smectite and illite contents are higher in the sandy shale/shaley portions of the Wells.

Table 1. Relative abundance of Clay minerals expressed as %Clay content in Well 1.

Depth	Kaolinite	Illite	Smectite	Chlorite	Inference
4550	73.68	15.78	10.53	0.00	S ₁ -U Low smectite, high continental influence
4850	100.00	0.00	0.00	0.00	
5270	85.00	0.00	5.00	10.00	
5450	63.64	0.00	0.00	36.36	
6105	42.80	2.55	29.37	26.07	V-Y High smectite, high deltaic basinal influence
6355	30.10	12.20	31.93	33.85	
7255	50.41	15.24	37.40	0.00	
7555	52.44	9.09	32.32	0.00	
8455	60.14	9.09	30.77	0.00	
9055	54.27	15.24	30.49	0.00	
9355	45.68	22.04	32.28	0.00	Z ₁ Low smectite, high continental influence
9655	69.76	2.25	27.98	0.00	
9955	29.35	4.52	37.42	28.71	

Table 2. Relative abundance of Clay minerals expressed as %Clay content in Well 2.

Depth	Kaolinite	Illite	Smectite	Chlorite	Inference
4030	70.00	0.00	17.50	12.50	S ₂ -U Low smectite, high continental influence
5230	98.50	0.00	1.50	0.00	
5700	85.72	8.57	5.71	0.00	
6315	96.63	0.00	3.37	0.00	
6735	58.72	9.88	31.40	0.00	V-Y High smectite, high deltaic basinal influence
7635	50.60	11.56	35.20	2.60	
8235	43.10	12.93	40.82	3.15	
8835	39.78	13.54	39.60	7.08	
9675	43.09	24.40	32.51	0.00	
10155	67.39	2.17	15.22	15.22	Z ₁ Low smectite, high continental influence
10515	29.01	5.33	38.33	27.33	

Table 3. Relative abundance of Clay minerals expressed as %Clay content in Well 5.

Depth	Kaolinite	Illite	Smectite	Chlorite	Inference
9880	52.81	4.34	32.65	10.00	Y High smectite, high deltaic basinal influence
10060	57.41	17.70	14.42	10.47	
10150	32.43	26.50	31.07	15.53	
10680	58.15	24.03	7.02	10.83	Z ₁ - Z ₀ Low smectite, high continental influence
10800	44.91	25.09	9.98	20.02	
11070	33.17	38.25	8.38	20.20	
11370	42.78	41.90	5.21	10.11	
11700	36.74	45.24	8.42	10.03	

The parameters (in percentages) of kaolinite, illite, smectite chlorite, fine sand, medium sand and coarse sand were subjected to Pearson correlation coefficients analysis (Tables 4-6). The significant positive

correlations (smectite to fine sand in Table 4; kaolinite to medium sand, illite to smectite, and smectite to silt / shale in Tables 5; and smectite to fine sand in Table 6 are not representative of all wells and therefore cannot be used for suggestive deductions.

Table 4. Pearson Correlations for relative abundance of Clay minerals expressed as %Clay content and %Grain sizes in Well 1(N=13).

	Kaolinite	Illite	Smectite	Chlorite	Shale/Silt	Fine Sand	Medium Sand	Coarse Sand
Kaolinite	1.000	-.235	-.659*	-.545	-.010	-.200	.119	-.157
Illite		1.000	.518	.154	.457	.109	-.421	.401
Smectite			1.000	.179	.679*	.590*	-.740**	.383
Chlorite				1.000	-.318	-.014	.099	.155
Shale/Silt					1.000	.370	-.823**	-.409
Fine Sand						1.000	-.711**	.537
Medium Sand							1.000	-.971**
Coarse Sand								1.000

* Correlation is significant at the 0.05 level (2-tailed).

** Correlation is significant at the 0.01 level (2-tailed).

Table 5. Pearson Correlations for relative abundance of Clay minerals expressed as %Clay content and %Grain sizes in Well 1(N=11).

	Kaolinite	Illite	Smectite	Chlorite	Shale/Silt	Fine Sand	Medium Sand	Coarse Sand
Kaolinite	1.000	-.632*	-.964**	-.462	-.734*	-.718	.959**	-.750
Illite		1.000	.654*	-.299	.540	.407	-.770*	.745
Smectite			1.000	.284	.639*	.763	-.931**	.664
Chlorite				1.000	.393	.248	-.544	.143
Shale/Silt					1.000	.591	-.879**	.387
Fine Sand						1.000	-.848**	-.392
Medium Sand							1.000	-.959**
Coarse Sand								1.000

* Correlation is significant at the 0.05 level (2-tailed).

** Correlation is significant at the 0.01 level (2-tailed).

Table 6. Pearson Correlations for relative abundance of Clay minerals expressed as %Clay content and %Grain sizes in Well 1(N=8).

	Kaolinite	Illite	Smectite	Chlorite	Shale/Silt	Fine Sand	Medium Sand	Coarse Sand	
Kaolinite	1.000	-.648	-.028	-.513	.361	-.151	-.286	1.000**	
Illite		1.000	-.680	.149	-.712*	-.516	.631	-1.000**	
Smectite			1.000	-.079	.389	.730*	-.392	-1.000**	
Chlorite				1.000	.419	.382	-.695	1.000**	
Shale/Silt					1.000	.734*	-.963**	-1.000**	
Fine Sand						1.000	-.842*	-1.000**	
Medium Sand							1.000	-1.000**	
Coarse Sand								1.000	

** Correlation is significant at the 0.01 level (2-tailed).

* Correlation is significant at the 0.05 level (2-tailed).

DISCUSSION

Kaolinite

In Well 1 and 2, the lithologic sandstone units ($S_1 - U$) contain the highest proportions of kaolinite (63.4 – 100%) with a mean of 84.15%. According to Pastouret et al (1978), the dominance of kaolinite in sediments in the Niger Delta sediments indicates the dominance of continental/terrestrial influence. The high proportion of kaolinite in these sediment shows the nearshore (coastal – upper neritic) depositional sites for these unit.

In the middle stratigraphic units ($V - Y$), the proportion of kaolinite is much lower (39.78 – 60.14% with a mean of 45.61%) indicating reduced continental influence. In the lowermost portion of the Umutu Wells, kaolinite constitute between 29.01 and 69.77% (mean 61.93%) of the total clay minerals. The interpretation of the kaolinite distribution pattern shows classification into three different zones representing near-shore influence in the lowermost portions (Z_1 and Z_0), deeper offshore ($V - Y$) basinal influence in the middle part and again near-shore/continental influence in the topmost zones ($S_2 - U$).

Smectite

Smectite contents of lithologic units ($S_1 - U$) ranges from 0.00 – 17.50% (mean=5.44%) of the clay contents while lithologic units ($V-Y$) and (Z_1 and Z_0) have 10-40.80% (mean=32.15%) and 8.38-37.42 (mean=16.60) respectively. While these smectite contents are low in ($S_1 - U$) and (Z_1 and Z_0) implying high continental influence, the $V-Y$ units in between have high values implying high basinal influence.

The occurrence of smectite at a stratigraphic horizons below the depth of 3km has been considered anomalous because the calculated corresponding geothermal temperature range between 100 – 120° C for the depth and also there is sufficient availability of potassium (Braide, 1993B).

Adeleye (1978) reported the occurrence of 14-15 A° smectite clays at 3.0 km depths in Shell's wells Ruta-1X, Mathin-1 and Ireti-1 on the western part of the Niger Delta, attributing this anomaly to sedimentation by rivers draining directly from the weathered basement of Southwestern Nigeria into the contemporary Guinea Sea as opposed to sedimentation brought into the delta by the Niger/Benue River drainage system.

Weaver (1978) reported that in the New Zealand geothermal area, the hydrothermal conversion of smectite to illite is more complex than normally encountered in sediments which have undergone burial diagenesis. He surmised that in area like the U.S. Gulf Coast the original sediments contain detrital smectite and beidellite, whereas in the geothermal areas the rocks are volcanic and are first altered to smectite before being converted to illite. In other words, the clay sedimentation facies are different prior to burial diagenesis.

Other studies include that of Rettike (1981) who observed that the occurrence of ordered illite/smectite from low temperature wells are attributable to provenance differences which gave lower-expandable illite/smectite a head start on diagenetic trends in sand associated shale.

Another Niger Delta study is that of Lambert-Aikhionbare and Shaw (1982), who believe that preservation of smectite in Agbada and Akata shales from the central part of the delta at burial temperatures in excess of what might be expected from accepted models of clay diagenesis reflect the under - compaction of the sediments arising from their rapid deposition. Dypvik (1983) discussed the possibility of classic source rock variation complicating clay mineral transformations. Veide and Nicot (1985) expound the view that diagenetic clay mineral composition is a function of pressure, temperature and chemical activity. Johnson and Reynolds (1986) suggest that both variation in provenance and climate can account for clay mineral variation. Jennings and Thompson (1986) support the idea that reaction rates are important in determining clay mineral assemblages at temperatures below 175°C.

Yau et al (1987) recognized smectite at depths that has been subjected to 200°C and interpreted its occurrence to be due to localized limited porosity.

As most clay are originally derived from the weathering of other minerals and volcanic material, climate and weathering conditions in general play a major role in determining the ultimate composition of shales (Millot 1970). In this regard, it can be assumed that the general composition of shales is in part determined by tectonic activity. For instance, it appears that the authigenic formation of clay minerals under marine conditions is at a maximum under periods of tectonic quiescence (Weaver 1978).

Shales deposited in the marine environment are therefore more abundant than those in other environment (Powers 1957). In a similar finding, Braide, 1993A, claimed that the anomalous occurrence of smectite at what could be termed 'prohibitive' depths for its existence points to a probable difference in provenance. He attributed this to be a case of different clay sedimentation facies in which smectite belonging to a different facies has its inherent characteristics.

Illite

The contents of Illite in lithologic units ($S_1 - U$) ranges from 0-15.78% (mean=3.04); 2.55-26.50% (mean=13.75%) for lithologic units V-Y and 2.17-38.25% (mean=20.97%) for (Z_1 and Z_0). The intermittent absence of illite in lithologic units ($S_1 - U$) suggests warm periods/events possibly due to periodic exposure.

Chlorite

Values ranging from 0.00-36.36 were obtained for chlorite contents in lithologic units ($S_1 - U$); 0.00-15.53 (mean=7.26) for lithologic units (V-U) and 0.00-28.71 (mean=15.83) for (Z_1 and Z_0). Chlorite is generally considered to be associated with high attitudes and physical weathering. It cannot persist in areas of intense chemical weathering because it is easily eliminated by hydrolysis (Biscaye, 1965). The intermittent absence of chlorite in each lithologic units is supportive of warm period/events and also suggestive of intense chemical weathering.

CONCLUSION

This study showed the distribution of clay minerals within the depth of 4550-10515ft below the surface of Umutu oil field. The pattern of distribution of the clay minerals are indicative of three different zones representing near-shore influence in the lowermost portions (Z_1 and Z_0), deeper offshore (V – Y) basinal influence in the middle part and another near-shore/continental influence in the topmost zones ($S_2 - U$). The regions could have experienced periodic exposures during warm seasons and intense chemical weathering.

This study also captured high occurrence of smectite which is anomalous for such depths.

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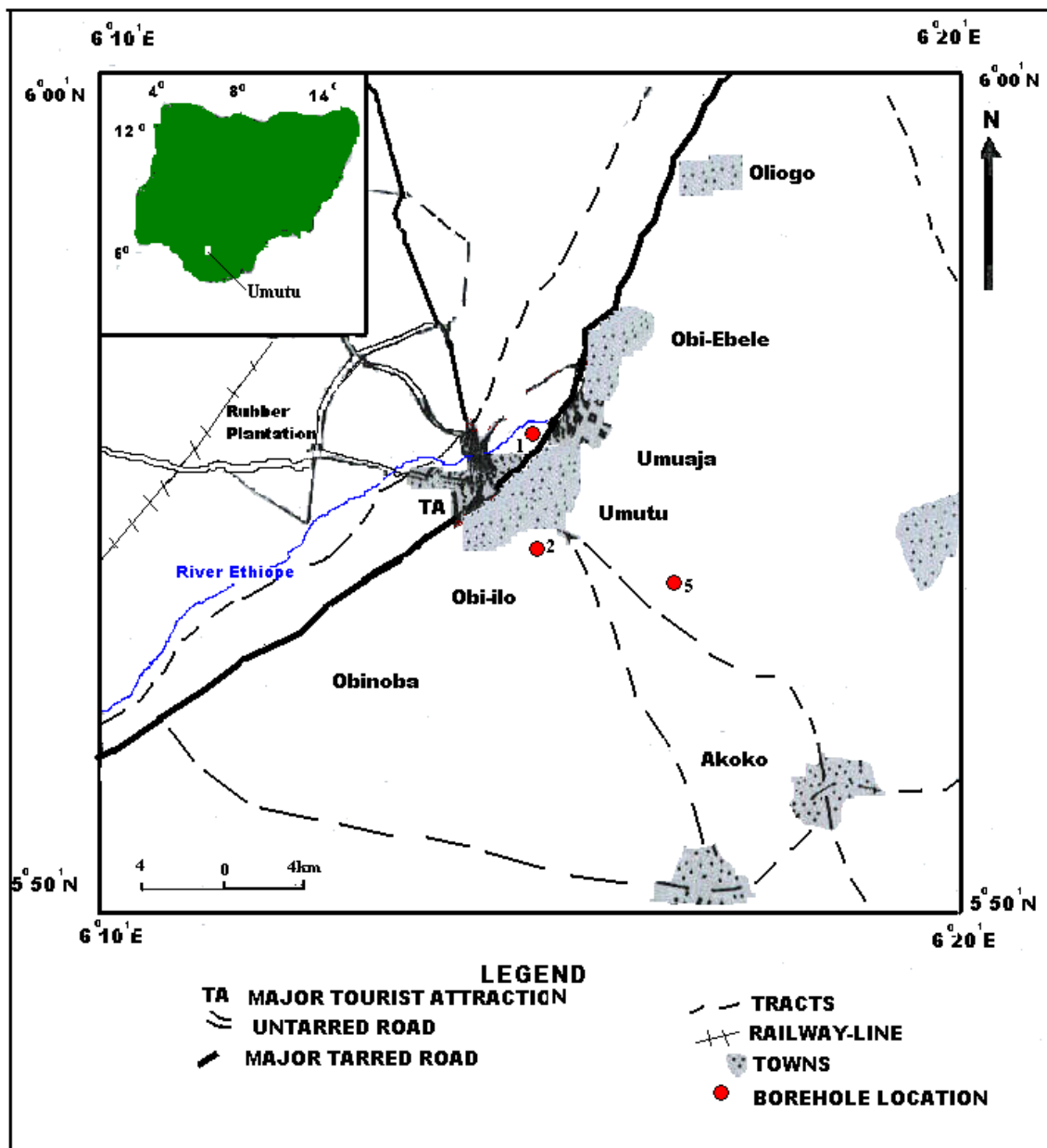


Figure 1 (colour online): Map of Umutu Area.

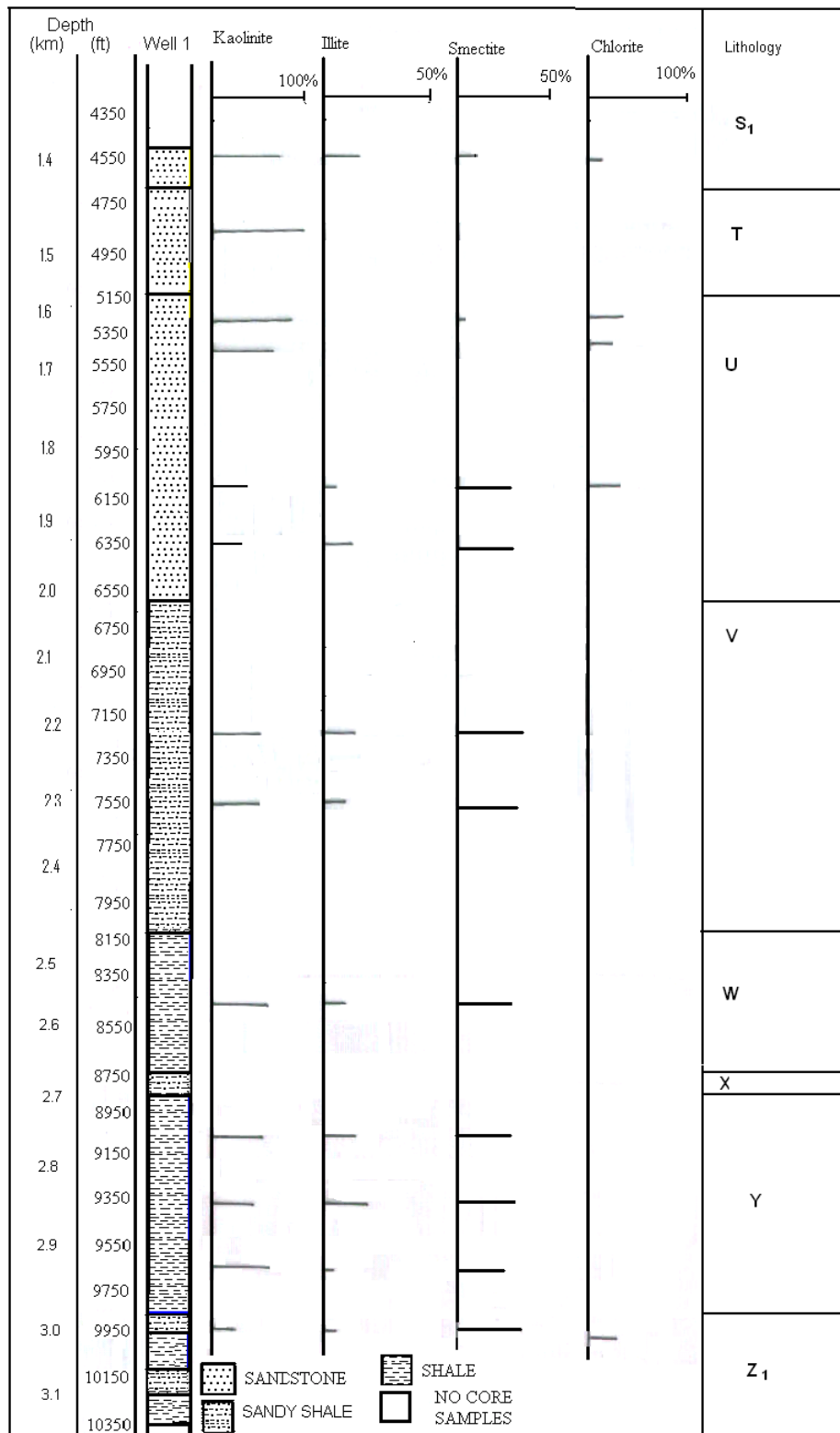


Figure 2. Relative Abundance of Clay Minerals expressed as %Clay Fraction (Well 1)

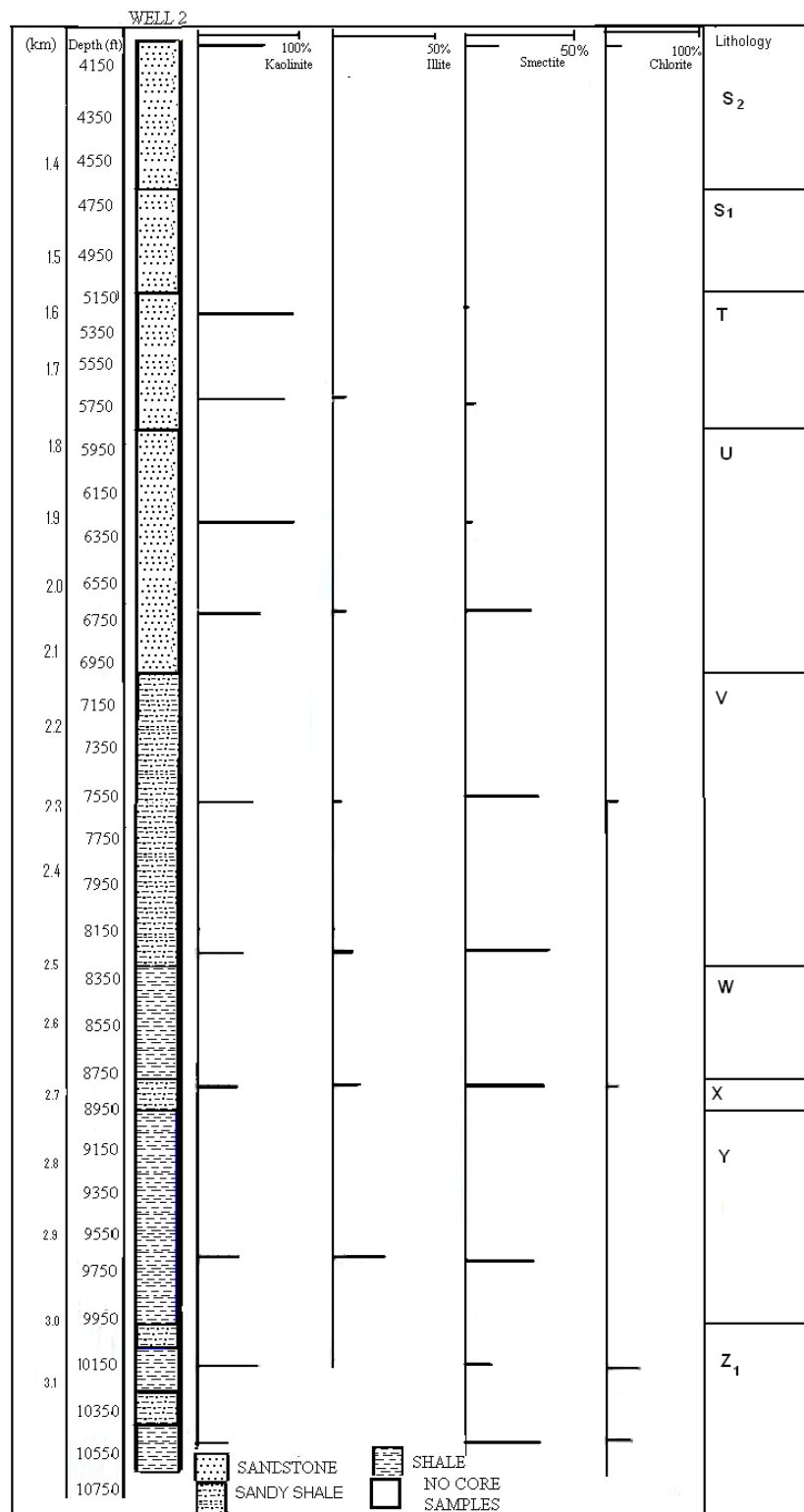


Figure 3. Relative Abundance of Clay Minerals expressed as %Clay Fraction in Umutu Well 2



Figure 4. Relative Abundance of Clay Minerals expressed as % Clay Fraction in Umutu Well 5.

THE USE OF VERY LOW FREQUENCY ELECTROMAGNETIC METHOD (VLF-EM) FOR HYDROGEOLOGIC INVESTIGATION IN PART OF THE FEDERAL UNIVERSITY OF TECHNOLOGY AKURE NIGERIA

M. O. Ofomola¹, J. O. Oseji¹ and G. U. Ozulu²

¹Department of Physics, Delta State University Abraka, Nigeria

²Department of Physical Sciences, Novena University Ogume, Delta State.

Email: ovirimerrious@yahoo.com

Abstract

The staff quarters of the Obanla campus of The Federal University of Technology, Akure has been investigated using the ABEM WADI VLF-EM instrument. Geophysical response obtained from the area which fall within the basement complex terrain depends on the rock types and the concentration of fractures in the study area. The result of the VLF-EM survey shows anomalies that were interpreted in terms of structures and lithology favourable to water saturation, thus allowing the hydrogeologic condition of the area to be determined.

Keyword: Very Low Frequency –Electromagnetic (VLF-EM), Aquifer, Groundwater Potential, Lineaments.

INTRODUCTION

Electromagnetic profiling is a widely used geophysical method in the delineation of basement regolith and location of fissured media and associated zones of deep weathering in crystalline terrains (Beeson and Jones, 1988; Olayinka and Olorunfemi, 1992). In many instances, the VLF-EM is used as reconnaissance tool to locate aquiferous zones such as fractures, faults and joints (Palacky et al, 1981; Omosuyi et al 2007; Mogaji and Oladapo, 2008).

The study area is the Obanla staff quarters of the Federal University of Technology Akure (FUTA). The site occupies an area of about 1km² and it lies within latitudes 7°18'21.3''N and 7°18'47.1''N (808000 and 808800 mN in the Universal Transverse Mercator scale, UTM), and longitudes 5°07'27.0''E and 5°08'19.2''E (734600 and 736200 mE in the Universal Transverse Mercator scale, UTM both in Datum 100 Minna – Nigeria and zone 31N 0°E to 6°E), Figures 1 and 2.

The study area is underlain by Basement Complex rocks of the southwestern Nigeria and groundwater in this environment is usually contained in the weathered and/or fractured basement rocks or alluvial deposits within flood plains as mentioned by some authors among whom are Wright (1992) and Olorunfemi and Fasuyi (1993).

In this work, geophysical mapping of the staff quarters was carried out using the VLF-EM prospecting technique with a view to identify anomalous areas, in relation to groundwater potential and ultimately recommend areas suitable for water wells.

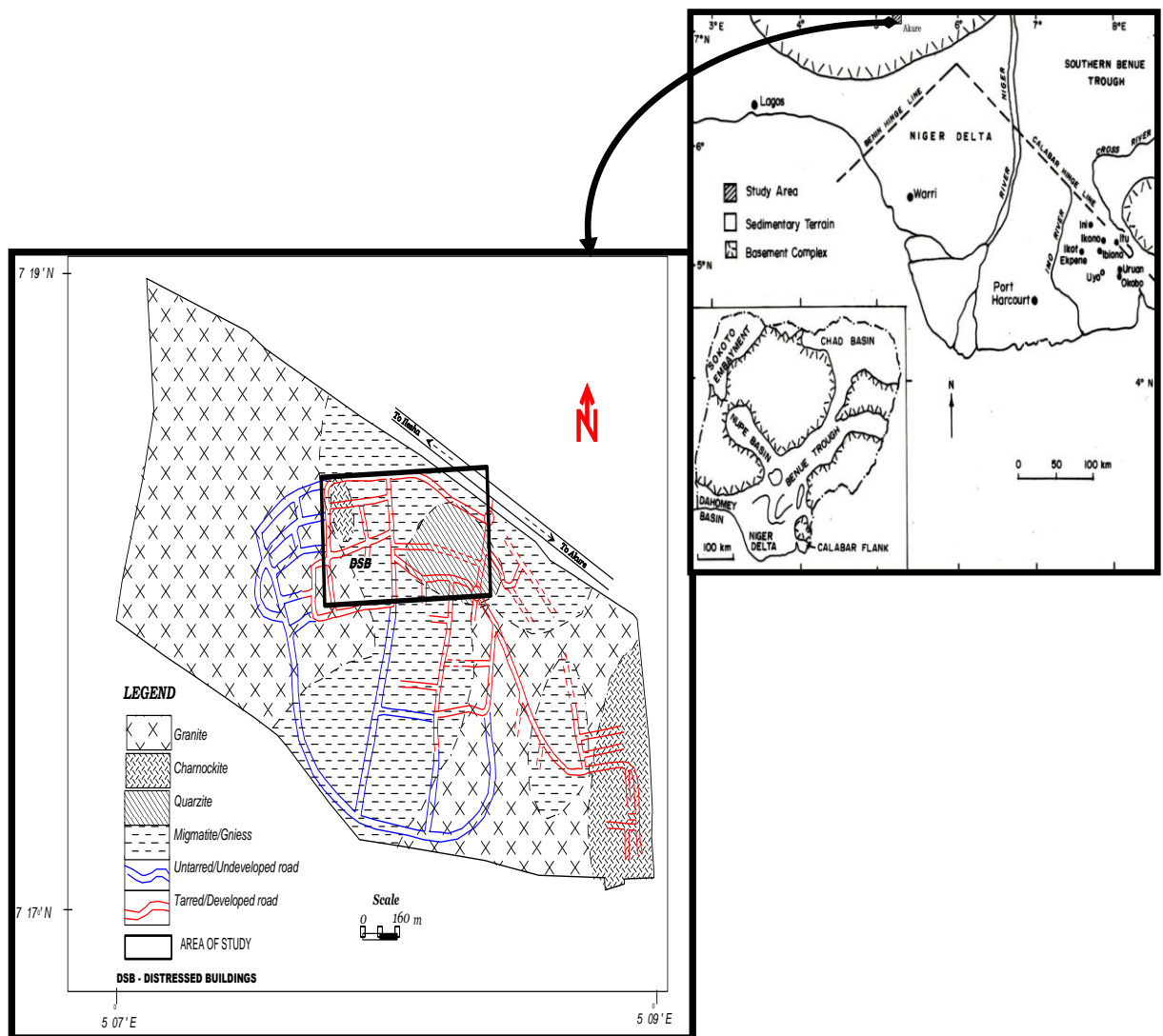


Fig. 1. (colour online): Location and Geological Map of the study area

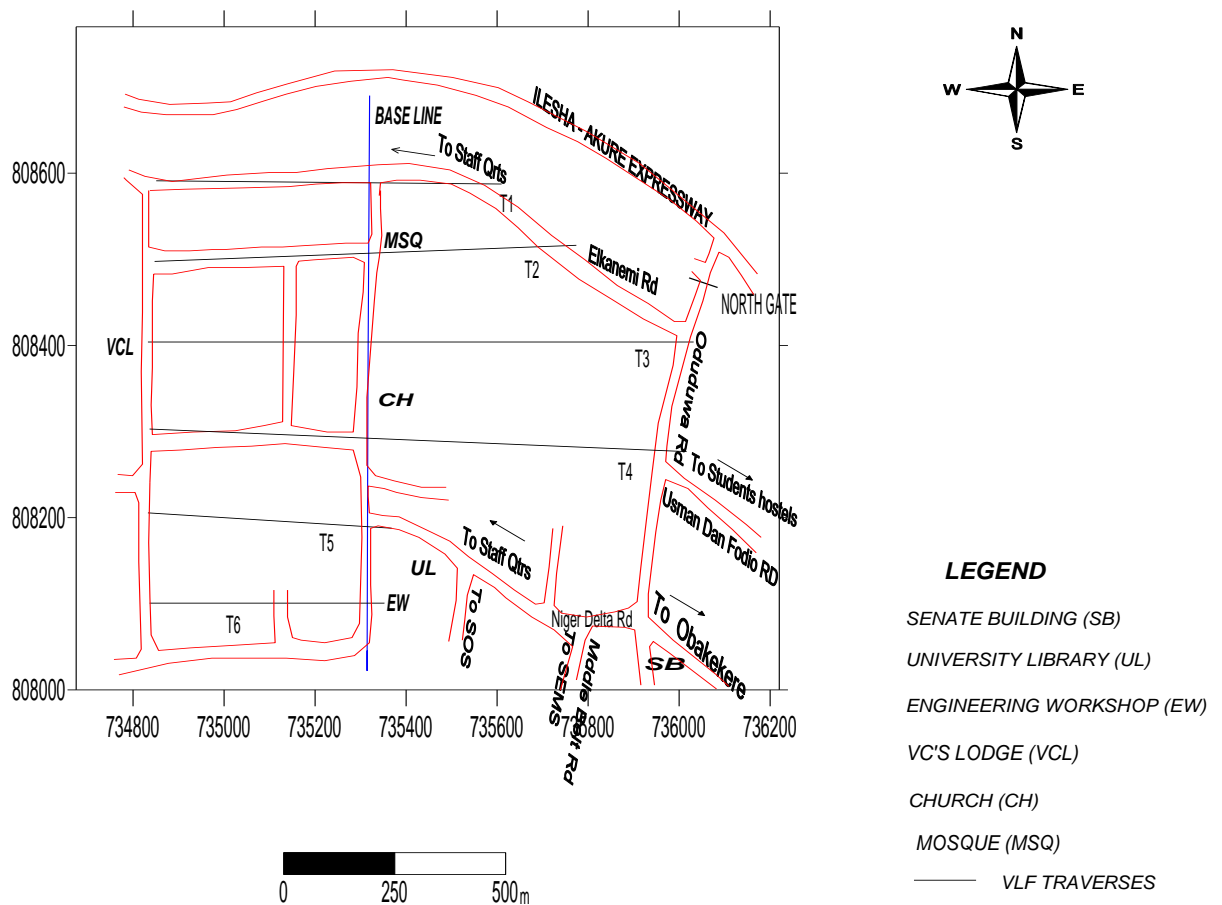


Fig. 2. (colour online): Geophysical Field Layout Map of the study area

LOCAL GEOLOGY

The study area is underlain by Precambrian rocks typical of the Basement Complex of southwestern Nigeria (Rahaman, 1976), comprising of undifferentiated granite, Charnokitic rocks, medium to coarse granite and Migmatite gneiss rocks and quartzites (Figure 1). The aquifer units in the area and other similar Basement complex environment are believed to be derived essentially from the weathered rocks (Bala and Onugba, 2001; Olayinka and Olayiwola, 2001). The weathered profile developed above the crystalline basement rocks in low latitude regions where the study area lies has been documented to comprise, from top to bottom, the soil layer, the saprolite (i.e., the product of the in situ chemical weathering of the bedrock), the saprock (fractured bedrock) and fresh bedrock (Olayinka and Olayiwola, 2001). The area as part of the Federal University of Technology Akure, lies within the tropical rain forest belt characterised by alternating wet and dry seasons with a mean annual rainfall of 2000mm. Humidity is relatively high during the wet season and low during the dry season. Temperature varies from 22°C to 29°C throughout the year (NIMET, 2007).

MATERIALS AND METHODS

The VLF-EM geophysical method is a quick and powerful tool for the study of shallow conducting lineament features in the near surface earth (Telford et al 1977). The method is based on measurement of

the secondary magnetic field induced in local conductors by primary electromagnetic fields generated by powerful military radio transmitter in the very low frequency range (15 – 25 KHz).

The instrument employed for this survey is the ABEM WADI, which measures the in-phase (Real) and quadrature (Imaginary) components of the induced vertical magnetic field as a percentage of the horizontal primary field.

The in-line profiling technique was employed for the VLF-EM using a station separation of 10m along six pre-cut traverses with traverse length varying from 570m to 1230m (Figure 2). In otherwords traverses were cut perpendicular to established geologic strike direction in the area and the transmitter signal direction. Inphase and quadrature values in percentages were plotted against station positions using the Microsoft Excel Package. Qualitatively, the varying amplitude from this anomaly profiles is a measure of the conductivity changes in the subsurface. Also, the Karous - Hjelt and Fraser filter (Pirttijarvi, 2004) package was used to perform Karous - Hjelt and Fraser filtering on the VLF – EM data in order to produce 2-D models along the traverses in the study area.

RESULTS AND DISCUSSIONS

The plots of filtered real (quadrature) components are presented as profiles and their corresponding Karous-Hjelt (K-H) pseudo sections are shown in Figures 3-8. The interpretation of both the profiles and pseudo sections was basically qualitative or semi quantitative. The anomaly inflections appear as peak positive anomalies and false VLF anomaly infections as negative anomalies (Reynolds, 1997) of the profiles.

The varying amplitude which is a measure of the anomaly changes in the subsurface vary from -54% to 55% in the study area, indicating variations in conductivities of the subsurface materials. From the default on the WADI equipment, the filtered real part will always show a positive peak above a conductor (Ofomola et al 2009).

Figure 3a shows the filtered real profile along traverse 1, anomalies with positive peaks amplitude on the filtered Real component. These anomalies include those at stations -150 (15W), 0 and 130 (13E). These points are zones of interest for viable groundwater in basement complex terrain. The corresponding K-H 2D model of the profile which is a measure of conductivity of the subsurface as a function of depth is also shown in figure 3b. This conductivity is shown in colour codes with response increasing from left to right (i.e. from negative to positive). Conductive features of varying degree of conductivity trending in different directions were delineated on the section, for instance, a conductive body of response of 20% is shown around 150 (15W). Similarly, between stations 0 and 130 (13E), another conductive feature of response of 15% all trending in the SW-NE directions is observed. Conductive bodies present on the section coinciding with the points already identified on the profile, as fractures/joints. The same process of qualitative interpretation was adopted for the remaining profiles and their corresponding 2D models generated as shown in figures 4 - 8.

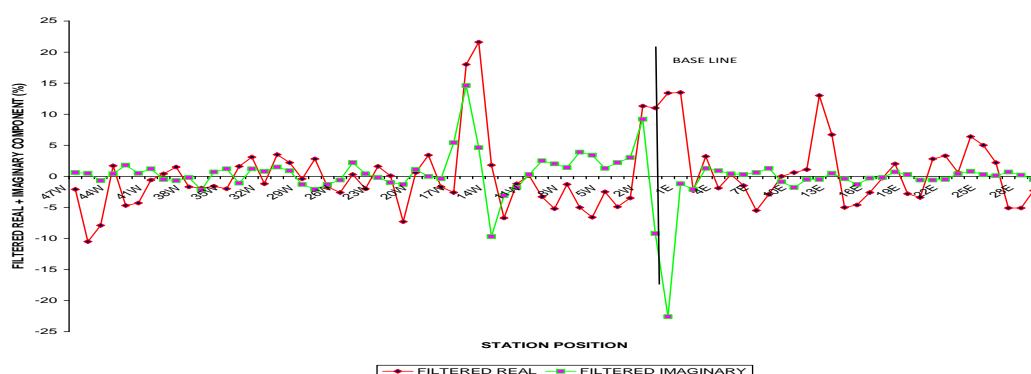


Fig 3a (colour online): VLF-EM profile along traverse 1

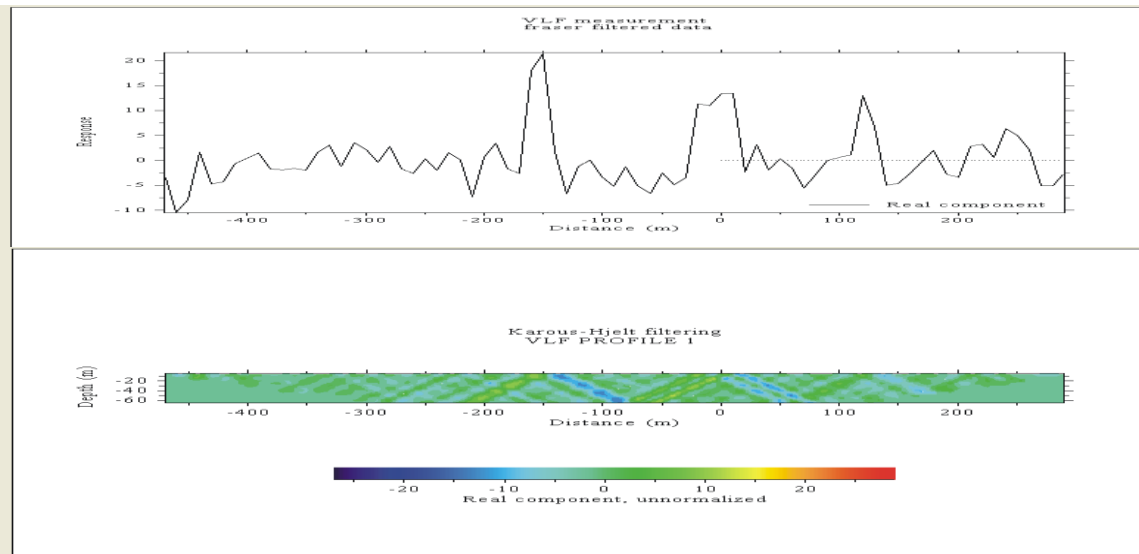


Fig 3b (colour online): Corresponding VLF-EM profile and 2D section along traverse 1 using KHFFilter

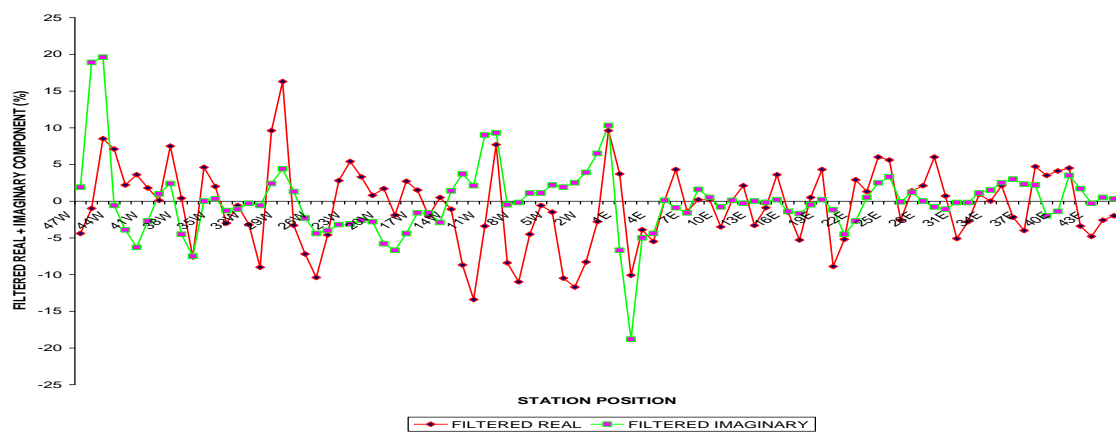


Fig 4a: VLF-EM profile along traverse 2

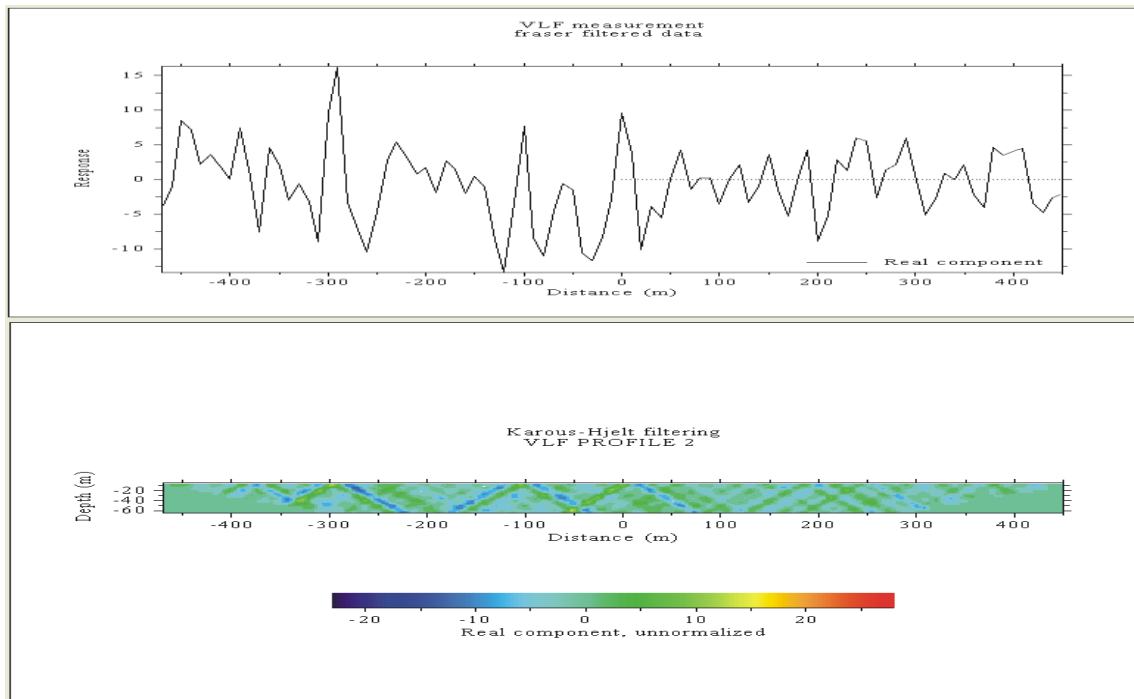


Fig 4b (colour online): Corresponding VLF-EM profile and 2D section along traverse 2 using KHFFilter

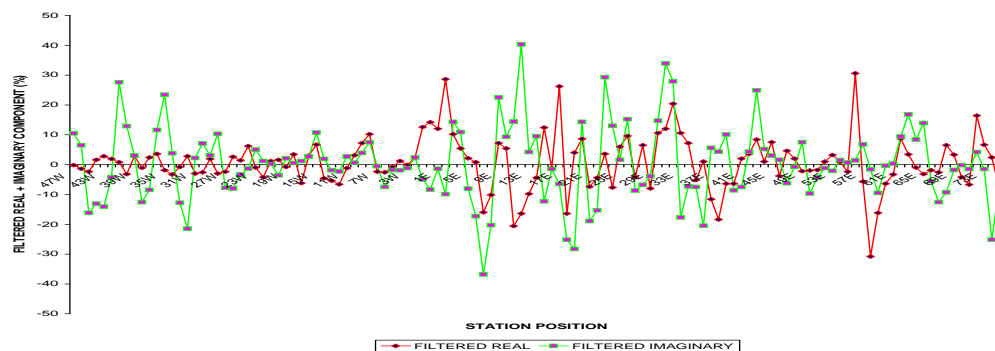


Fig 5a (colour online): VLF-EM profile along traverse 3

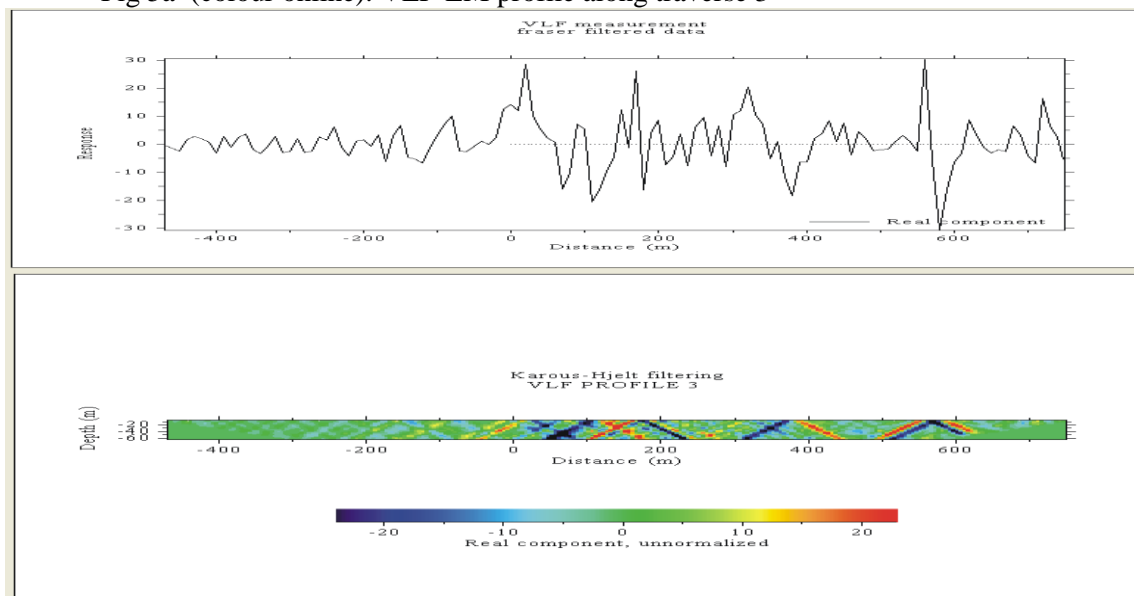


Fig 5b (colour online): Corresponding VLF-EM profile and 2D section along traverse 3 using KHFFilter

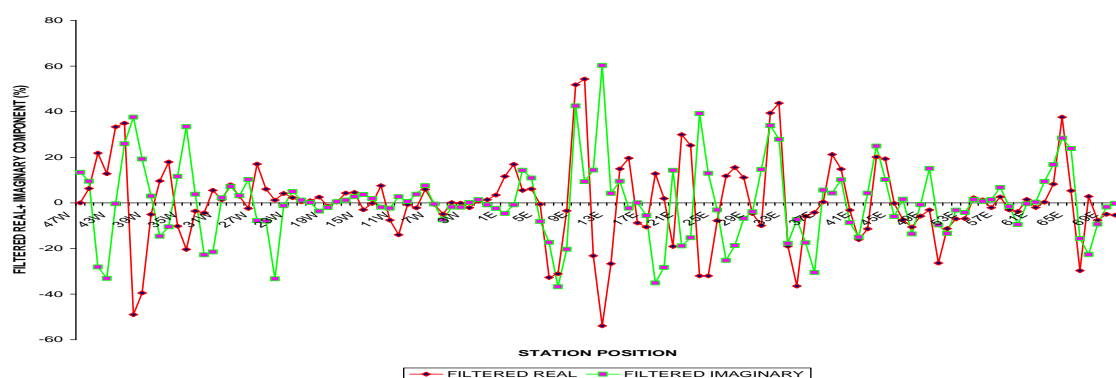


Fig 6a (colour online): VLF-EM profile along traverse 4

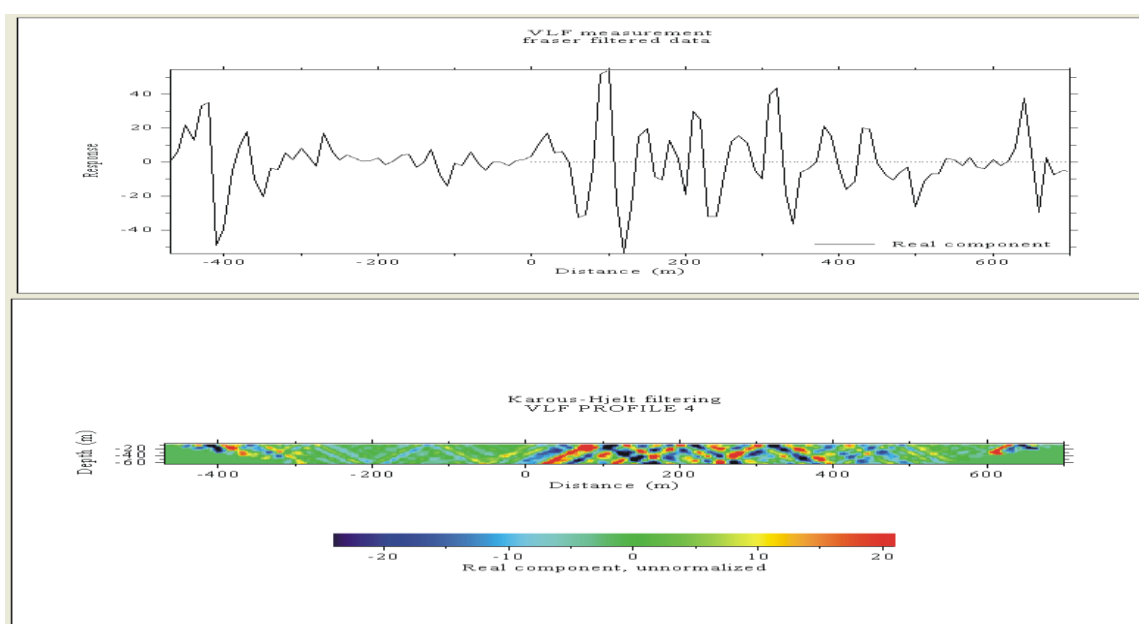


Fig 6b (colour online): Corresponding VLF-EM profile and 2D section along traverse 4 using KHFFilter

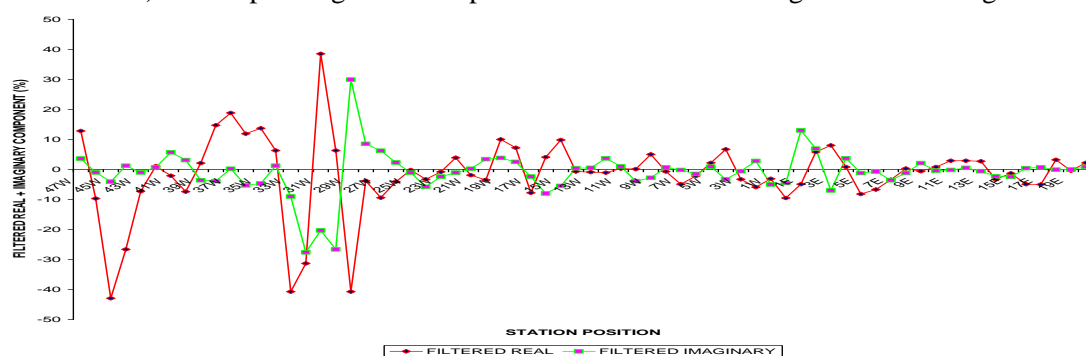


Fig 7a (colour online): VLF-EM profile along traverse 5

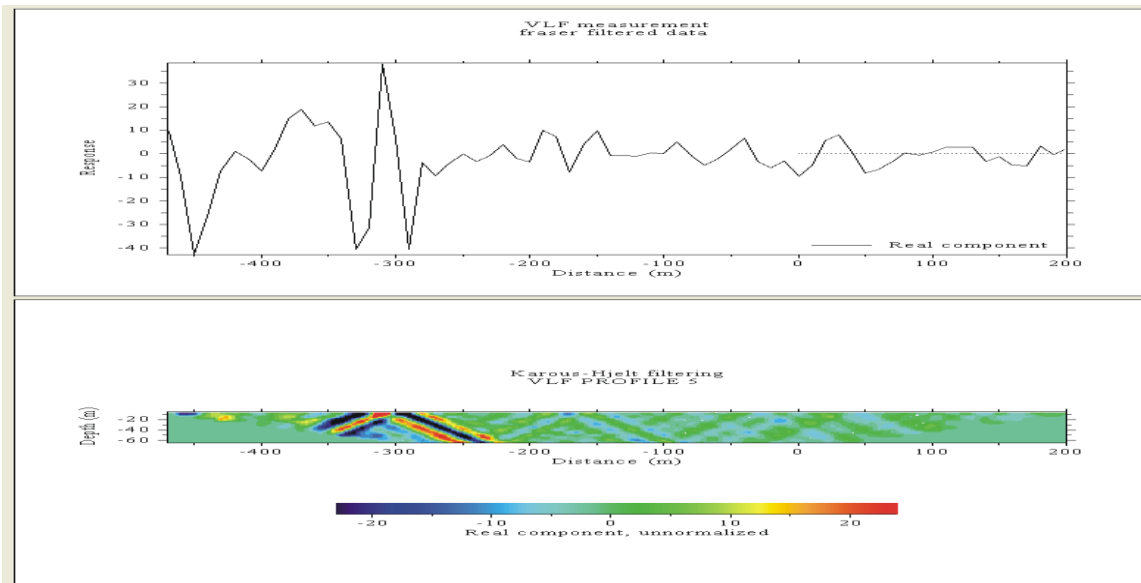


Fig 7b (colour online): Corresponding VLF-EM profile and 2D section along traverse 5 using KHFFilter

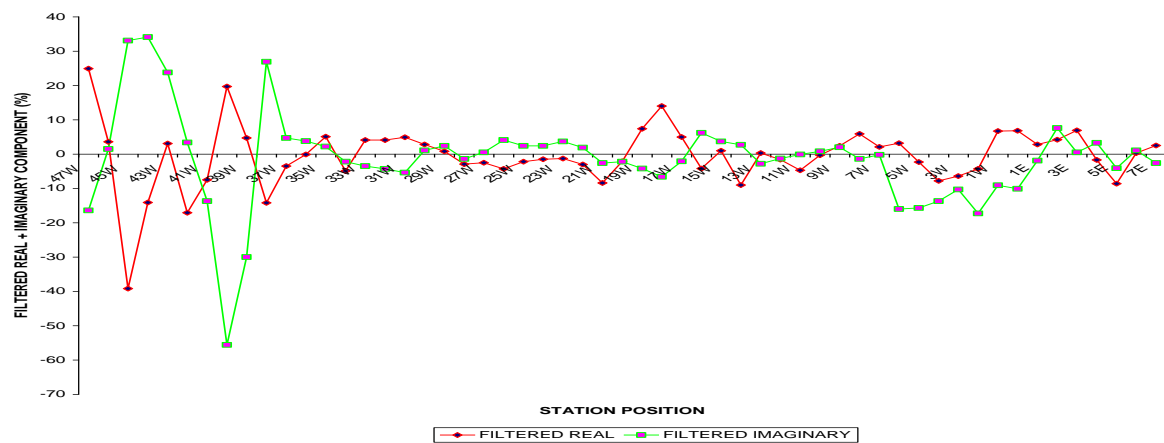


Fig 8a (colour online): VLF-EM profile along traverse 6

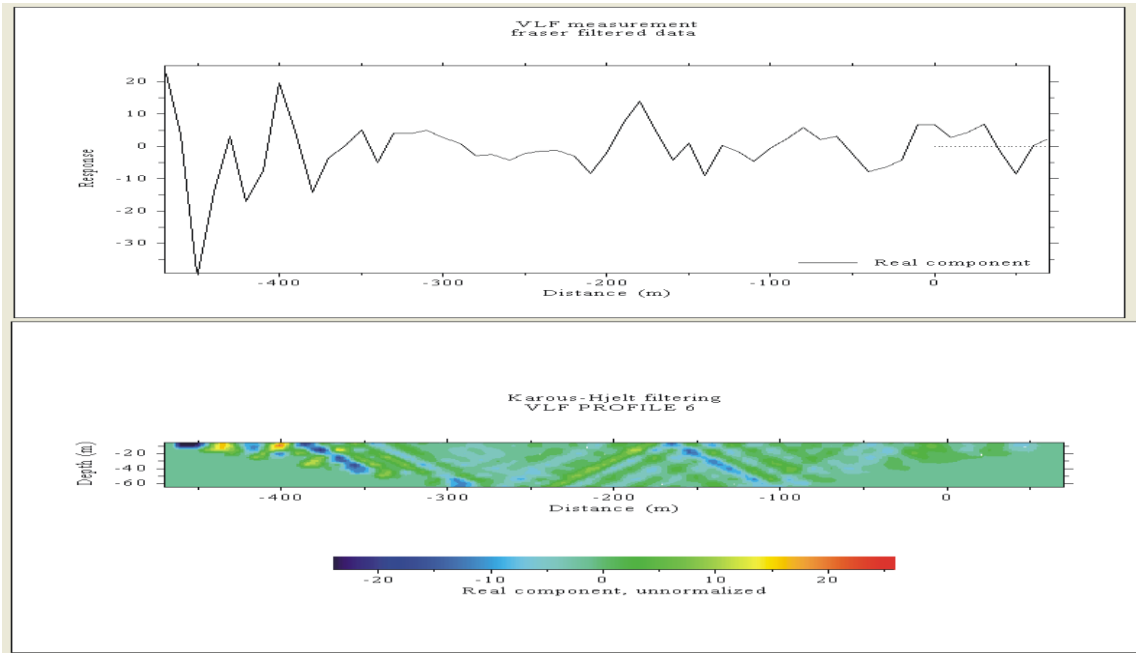


Fig 8b (colour online): Corresponding VLF-EM profile and 2D section along traverse 6 using KHFFilter

In addition, a lineament map (Fig 9) of the study area was also generated. This map shows the concentration and azimuth direction of the lineaments in the study area. Conductive features of the pseudo-sections from the KHFFILT were marked and their position and orientation drawn on a profile line of traverses one to six using the same scale. From the map, the area with high concentration of lineaments correlates with the conductive zones from the profiles (e.g the eastern flank of traverses 3 and 4, and the south-western part of the study area). These areas of high concentration of lineaments are zones of interest in groundwater abstraction in basement complex terrain.

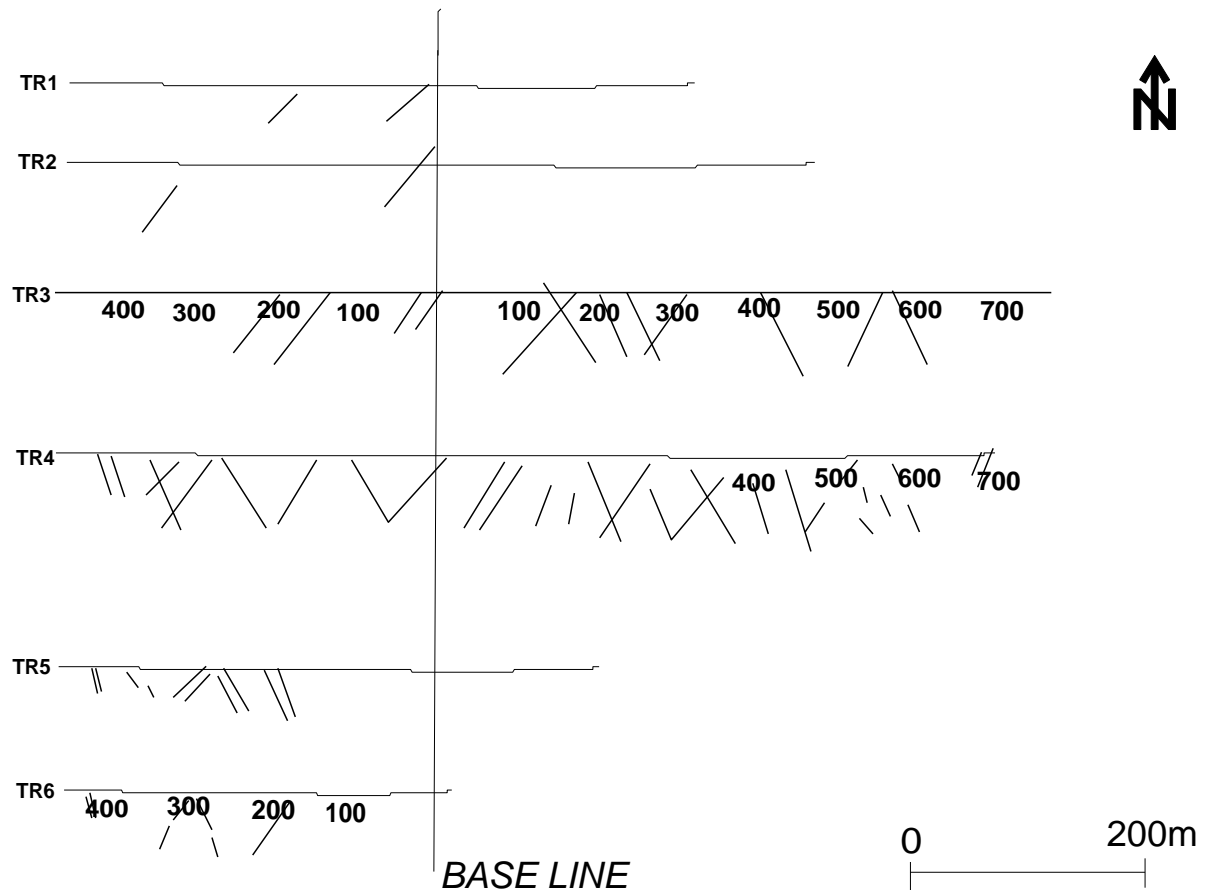


Figure 9: Lineament map of the study area

CONCLUSIONS

The VLF-EM method has greatly assisted in evaluating groundwater potential of Obanla staff quarters of The Federal University of Technology, Akure. Site with high electromagnetic anomaly (high positive filtered real anomaly) can be expected to be weathered basement/fractured zones, implying locations suitable for the development of groundwater resources. Also zones around traverses 3 and 4 as shown in the generated lineament map have been identified as the most probable areas for groundwater development in the area due to the high concentration of fractures in that area. This study serves as a guide for future follow-up using other relevant geophysical methods

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THE USE OF EARTHCLAYS IN WATER PURIFICATION

Umudi, E. Q.*, Adaikpoh, E. O.**, Okoh, B. E.* and Awatefe, K. J.*

*Department Of Chemistry, College Of Education, Agbor. Delta State Nigeria.

** Department of Geology, Delta State University, Abraka. Delta State, Nigeria.

*e-mail: ese.umudi@yahoo.com

Abstract

The geochemical and mineralogical analyses of clays obtained from Otor Edo (OT) and Abbi (BB) in Nigeria were studied using X-ray diffractometer and Atomic Absorption Spectrophotometer. The mineralogical analysis showed the clay mineral contents of OT as kaolinite and illite while BB contains kaolinite, simectite, illite and mixed layer. Geochemical analysis show that the hydrated alumina silicate are associated with other elements like Fe, Na, Mg, Ca, Ti and K. During purification, Earthclays fortified (i.e. Pebble: clay = 1:2 w/w) gave optimum water purification. BB gave the lower percolation rate studies using 3 columns. The overall performance of clays in Brewery wastewater treatment was good. The percentage removal of total Bacteria was above 91% with BB giving a higher pollutant removal. The pH, total solid, Dissolved Oxygen and Chemical oxygen Demand were monitored for fourteen days and there was reduction. Clays BB and OT achieved complete removal of the metals. This is a simple, reliable low-cost, low energy consuming and technologically simple decentralized waste water purifying system.

KEYWORDS: Earthclays, purification, mineralogical content, pollutants.

INTRODUCTION

Every human being deserves safe drinking water, but much of the world population do not have access to it. Water is vital to life as it is a fundamental requirement for the survival, well being and social-economic development of all humanity. It is a solvent medium, transport medium and catalyst in nearly all chemical reactions occurring in the environment (Ademoroti, 1996). The foremost position of water in man's daily operation has left him with no option other than to recycle such water for reuse. The mammoth demand for clean water has always been met by tapping water from its aquifers, one of the earth's main deposit of fresh water, but the sudden realization that the supply is not inexhaustible has awakened the interest of researchers to devise sustainable means of water/wastewater treatment for reuse.

Two distinctive properties of clay that render them technologically useful are plasticity when wet and their extremely fine crystals often colloid in size and platy in shape. The flattened (platy) shape of clay particles together with peculiarities of the crystal structure give opportunity for adsorbing ions in a variety of ways. Some ions are held to broken bonds on the edges of the flakes while others are held to the flat surface (Farmer, and Motle, 1966). Some make their way into spaces between the layers of the crystal structure and some into the crystal itself to take the place of one of its constituents. This depends on the kind of ions, and the kind of clay minerals present (Grim, 1968).

This paper is aimed at designing a simple reliable low cost, low energy consuming and technologically simple decentralized wastewater purifying system using different clay minerals fortified with stone pebbles to overcome the problem of soil impermeability to water, which has restricted its use as percolator material.

MATERIALS AND METHODS

Clays were collected from Otor-Edo (OT) and Abbi (BB) (Delta State), air-dried pulverized with wooden mortar and sieved with 0.05 size filter. Stone pebbles were collected from sharp sand dug from Ethiopia river, washed thoroughly under tap water and air dried.

Brewery (Wastewater)

The wastewater used was obtained from a brewery in Lagos. The various sources of the wastewater were from malt preparation and fermentation process. These were formed during barley washing and steeping and bottling of beer products. The colour produced was yellowish-brown and displayed a tendency towards foaming and putrefaction. All the effluents were discharged from the brewing plant, fermentation and storage cellars, bottling and washing plants and equipment washing process. The amount of waste generated daily was about 19.1×10^4 litres (42×10^3 gallons). They were collected in 10 litres plastic containers at 2 hours interval over a period of 12 hours starting at 6.00am in the morning and ending at 6.00pm. A composite sample was taken. pH determinations were carried out on the field. Samples were analysed for Turbidity, Suspended Solids, Total Solids, Dissolved Oxygen (DO), Biochemical Oxygen Demand (BOD); Chemical Oxygen Demand (COD), Ammonia – Nitrogen ($\text{NH}_4^+\text{-N}$), Total Kjeldahl Nitrogen (TKN). Nitrate-Nitrogen (NO_3^-N), Total bacteria and Heavy metals as recommended by Ademoroti (1996).

Mineralogical Analysis using X-ray diffractometer and Catio Exchange Capacity (CEC) using Black (1968), Performance Efficiency and Perculation Studies were according to APHA (1985).

RESULTS AND DISCUSSION

Table I: Mineralogical Analyses Clay Minerals

Clay Minerals	OT	BB
Saponite	Nil	Nil
Smectite	Nil	8.3
Chlorite	Nil	Nil
Illite	10.1	12.4
Mixed layer Illite-Montronillonite	Nil	15.4
Kaolinite	58.2	39.4
Quartz	27.5	24.5
Hematite	4.3	Nil

KEY: OT - Otor-Edo, BB - Abbi

Table 2: Results of Geochemical Analysis

Metal oxide %	OT	BB
SiO_2	43.37	44.44
Al_2O_3	38.60	38.80
Fe_2O_3	6.61	7.67
Na_2O	1.97	0.89
MgO	1.93	1.82
K_2O	2.69	1.68
TiO_2	0.99	0.98
CaO	0.31	0.21

Table 3: Cation Exchange Capacity

Clay Sample	CEC (m/kg)
OT	71.00
BB	62.00

Table 4: Results of Raw and Treated Brewery wastewater (Using Single Column)

Parameter	Units	Raw Sample	Results after Treatment	
			OT	BB
pH	-	6.31	7.80	7.60
Turbidity	NTU	200.00	1.2	1.1
TS	mg/L	300.00	7.50	6.80
DS	“	101.00	6.70	5.01
SS	“	190.00	0.82	0.87
DO	“	0.61	4.70	4.50
COD	“	710.00	48.00	40.20
BOD	“	240.00	16.00	14.00
TKN	“	17.50	3.10	3.00
NH ₄ ⁺ -N	“	16.20	1.01	1.02
NO ₃ -N	“	4.20	1.40	1.48
Total Bacteria	Count/100ml	4.0 x 10 ⁸	2.1 x 10 ⁶	1.3 x 10 ⁶

Table 5: Results of Raw and Treated Brewery Wastewater (Using three Columns)

Parameters	Units	Raw Sample	Results after Treatment	
			OT	BB
pH	-	6.31	8.00	8.20
Turbidity	NTU	200.00	ND	ND
TS	mg/L	300.00	1.9	ND
DS	“	101.00	2.00	1.80
SS	“	190.00	ND	ND
DO	“	0.61	9.00	8.50
COD	“	710.00	9.20	9.10
BOD	“	240.00	6.00	5.32
TKN	“	17.50	0.10	0.11
NH ₄ ⁺ -N	“	16.20	0.06	0.03
NO ₃ -N	“	4.20	0.50	0.41
Total Bacteria	Count/100ml	4.0 x 10 ⁸	7.2 x 10 ⁴	6.9 x 10 ⁴

Table 6: Metal Ion Adsorption

Metal Ions	Units	Raw Sample	Results after Treatment	
			OT	BB
Cr	mg/L	0.040	-	-
Hg	“	ND	ND	-
Pb	“	0.430	-	-
Cd	“	<0.001	-	-
Fe	“	1.41	0.32	0.10

Table 7: Clay Ratio

Pebble Clay ratio	Raw Sample	Results after Treatment	
		OT	BB
1.2	100	6.00	8.00
1.3	100	12.00	16.00
1.4	100	10.00	12.00
2.1	100	15.00	13.00

Table 8: Results of Percolation Rate Studies Pebbles: Clay Ratio 1:2)

Clay Type	Mean Percolation Rate (m ³ /s)	
OT	To obtain first drop 2.90 x 10 ⁷	To collect 100ml 2.01 x 10 ⁷
BB	2.00 x 10 ⁷	1.90 x 10 ⁷

The kaolin group is the predominant the clay mineral OT 27.50% and BB 24.50%. The pH from each media was slightly alkaline due to cation removal from clay structure. TS reduction was about 86% with clay acting as inert medium for a fixed surface for microbial attachment and growth, reducing total solid content (Dinges, 1978). The BOD and COD which are indicators of pollution shows above 90% reduction using the 3 columns. The results of Nitrogen removal were about 60% removal in line with Kamppi (1971). Total Bacteria removal was well above 90% (Brown, 1972).

The mineralogical composition, chemical co-ordination characteristics and high surface contributes to their adsorptive power (Oladoja, 2003).

Conclusion

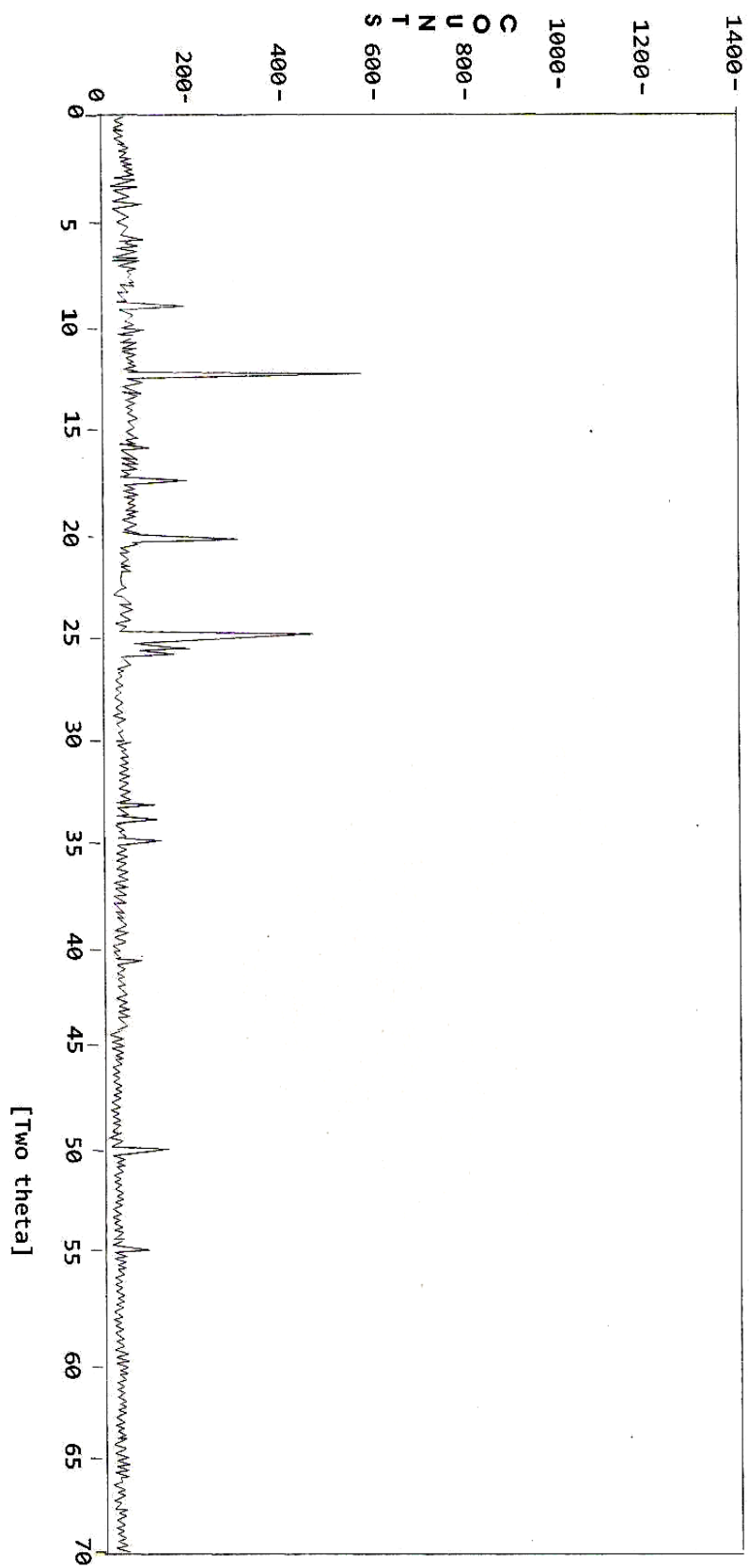
The three (3) column method gave a significant reduction of the pollution parameters studied and treatment efficiency of clays is directly related to their mineralogical assemblage. This study revealed that clays have high potential in wastewater purification.

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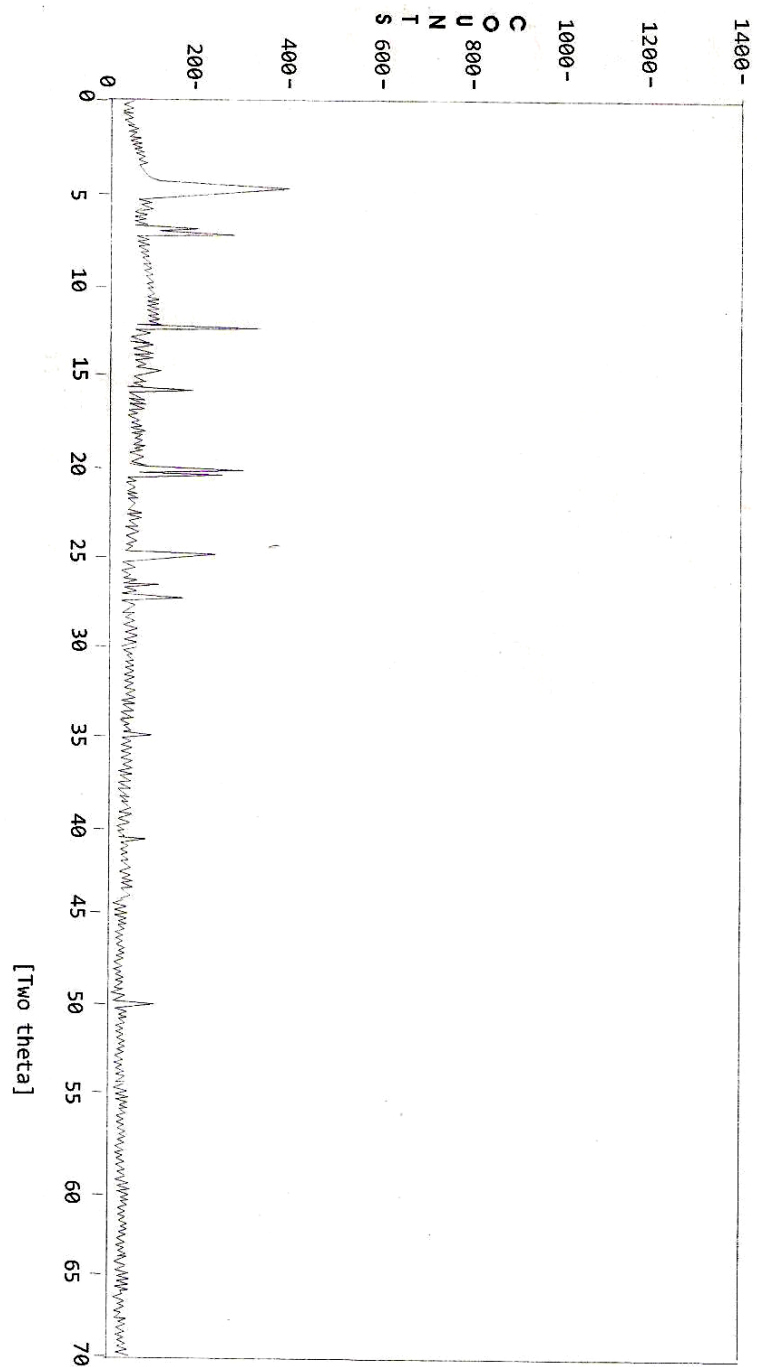
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File:C:\User_daf\queen\au_01_06.cpi Date 28 Feb 2007 Comment:queen Faka/Clay type Soil OT



File: C:\User_dat Queen\Qu_01_06.cpi Date 23 Feb 2007 Comment: Queen Faka/Clay type Soil BB



CRITICAL PROPERTIES AND ACENTRIC FACTORS OF FATTY ACIDS BY GROUP CONTRIBUTION METHODS

K. O. Monago^{a*} and C. Otobrise^{b*}

^aDepartment of Pure and Industrial Chemistry, University of Port-Harcourt, Port-Harcourt

^bDepartment of Chemistry, Delta State University, Abraka, Nigeria.

^{b*}E-mail: otocharles@yahoo.com

^{b*}Phone: 08038930023

Abstract

Some Group Contribution methods for the prediction of critical temperature, critical pressure and acentric factor of fatty acids were considered. The properties estimated by the selected methods, were tested using the Peng-Robinson equation of state to predict the liquid densities of fatty acids and compared with experimental values. The results show a reasonable agreement between estimated and experimentally determined P-V-T properties, with a minimum average percentage deviation of 8.10%. The results also show that with increase in the number of carbon atoms, critical temperature and acentric factor increases, while the reverse is the case for critical pressure. Finally, the results suggest that the method of Constantinou and Gani is best for estimating critical properties of fatty acids and that the Peng-Robinson equation of state is capable of predicting liquid densities with quality comparable with results obtained from experimental P-V-T data.

Keywords: critical temperature, critical pressure, acentric factor, fatty acids.

Introduction

The knowledge of the critical properties of organic compounds is required to implement many equation of state (EOS) methods; which in turn lead to interesting thermodynamic properties that are required in the design and analysis of process plants. Finding reliable experimental values of these properties is not always possible. Critical temperatures higher than 750K cannot be experimentally determined accurately [1]. This is due to the fact that many organic compounds are thermally labile, decomposing at temperature near or below their critical temperature.

In design, the screening of alternatives depends on fast, yet accurate, evaluation of properties for a large number of pure compounds and mixtures [2,3]. Therefore, in computer aided process and product design, simple, efficient and reliable methods for the estimation of properties of organic compounds from their molecular structure are essential for the analysis and design of products and processes. A case-in-point where property estimation methods can lead to improved processing techniques is the vegetable oil industry. Here, the triglycerides constituting the major components of the oils are labile molecules with unknown critical properties, thus preventing the use of modern separation techniques in the vegetable oil processing industry.

For the estimation of physical and thermodynamic properties of pure compounds, Group Contribution methods are the most widely used [4-9]. In these methods, the property of a compound is estimated as a summation of the contributions of simple first-order groups which can occur in the molecular structure. When furnished with substantial information about the molecular structure of a compound, the accuracy in estimating its properties is evidently improved.

A Group Contribution method uses the principle that some simple aspects of the structures of chemical components are always the same in many different molecules [3]. The smallest common constituents are the atoms and the bonds. By using group or atom properties, the physical properties of pure compounds and mixtures can be predicted. This reduces the number of needed data dramatically. Instead of needing to know the properties of thousands or millions of components, only data for a few dozens or few hundreds of groups have to be known.

The objective of this study was to predict or estimate, with good accuracy, the critical properties and acentric factors of fatty acids. A data base of this sort will ensure the selection of the most economic separation technique from competing alternatives; serve as a basis in the analysis and design of plants for separation and purification of vegetable oils and aid the selection of the optimum process conditions.

Pure Component Constants

Critical properties such as critical temperature, pressure and volume, as well as acentric factor represent four widely used pure-component constants [10]. Critical temperature has been defined as the temperature above which a gas cannot be liquefied by isothermal compression alone. The maximum pressure required for liquefaction at this temperature is the critical pressure. The critical volume is the volume occupied by one mole of the substance at its critical temperature and pressure [1]. The acentric factor is a measure of the anisotropy in the intermolecular force potentials of the molecules of interest, and is defined as

$$\omega = -1 - \log_{10} \left[P^{\sigma} \frac{(T = 0.7T_c)}{P_c} \right] \quad (1)$$

Where ω is the acentric factor; P^{σ} is the vapour pressure in bar; T_b is the normal boiling temperature in kelvin; T_c and P_c are the critical temperatures and pressures in Kelvin and bar, respectively.

Methods for the Estimation of Critical Properties and Acentric Factors

The Method of Ambrose

Ambrose [6] described an incremental method of correlating critical temperature; it is based on the quantity

$$X = \frac{T_b}{T_c - T_b} \quad (2)$$

The route by which an unknown critical temperature may be calculated from the boiling temperature and an estimated value of X is given by the correlation $X = a + \Sigma \Delta_X$

Where $a = 1.242$ for all compounds except perfluorocarbons and monohydrogen substituted perfluorocarbons, and $\Sigma \Delta_X$ is the sum of the increments for each atom or group in the molecule as stipulated by Ambrose [6].

The Method of Lydersen

This estimation method employs structural contributions to estimate T_c , P_c and V_c . The relations are;

$$T_c = T_b \left[0.567 + \Sigma \Delta_T - (\Sigma \Delta_T)^2 \right]^{-1} \quad (4)$$

$$P_c = M (0.34 + \Sigma \Delta_p)^{-2} \quad (5)$$

$$V_c = 40 + \Sigma \Delta_v \quad (6)$$

The Δ quantities are evaluated by summing contributions for various atoms or groups of atoms. A table of Lydersen's critical property increments for various atoms or groups can be readily sourced [5, 10, 11].

The Method of Constantinou and Gani

Constantinou and Gani [9], proposed a new additive property estimation method which is based on conjugation operators and applicable to organic compounds. This method which does not require the normal boiling temperature of the compound, has the advantage of capturing the differences among isomers and leading to improved accuracy in the prediction of physical properties. However, the generation and enumeration of conjugate forms is a non trivial issue and requires a symbolic computing environment [12].

Property estimation by the method of Constantinou and Gani [9] is done at two levels, first-order and second-order. The correlations for critical temperature and pressure are stated as:

$$T_c = 181.128 \ln \left[\sum_j N_i C_i + W \sum_j M_j D_j \right] \quad (7)$$

$$P_c = 1.3705 + \frac{1}{\left(0.100220 + \sum_i N_i C_i + W \sum_j M_j D_j \right)^2} \quad (8)$$

Where C_i is the contribution of the first-order group type-i, which occurs N_i times and D_j is the contribution of the second-order group type-j, which occurs M_j times in a compound. The constant W is assigned the value of unity in compounds that require second-order estimations and zero in compounds requiring only first-order estimations.

The Lee-Kesler Equation for Predicting Acentric Factor

The vapour-pressure equation proposed by Lee and Kesler [13] is a corresponding states expression which relates the reduced vapour pressure $\left(\frac{P^\sigma}{P_c} \right)$ with the reduced temperature $\left(\frac{T}{T_c} \right)$ and the acentric factor ω .

When it is applied to Eq. (1), the corresponding expression for acentric factor is

$$\omega = \frac{\alpha}{\beta}$$

(9)

$$\alpha = -\ln P_c - 5.92714 + 6.09648 T_r^{-1} + 1.28862 \ln T_r - 0.169347 T_r^6 \quad (10)$$

$$\beta = 15.2518 - 15.6875 T_r^{-1} - 13.4721 \ln T_r + 0.43577 T_r^6 \quad (11)$$

$$T_r = \frac{T_b}{T_c} \quad (12)$$

Peng-Robinson Equation of State

The Peng-Robinson EOS was developed in 1976 [14]. Amongst other characteristics, its parameters are expressible in terms of the critical properties and the acentric factor. The Peng-Robinson EOS may be written:

$$P = \frac{RT}{v-b} - \frac{a}{(v+t_1 b)(v+t_2 b)} \quad (13)$$

Where P is the total pressure of the system, R is the gas constant, T is the absolute temperature of the system, V is the molar volume, a is the attractive parameter and b is the repulsive parameter. Also,

$$t_1 = 1 + \sqrt{2} \quad (14)$$

$$t_2 = 1 - \sqrt{2} \quad (15)$$

$$a = a_c \alpha \quad (16)$$

$$a_c = 0.457235 \frac{(RT_c)^2}{P_c} \quad (17)$$

$$\alpha^{0.5} = 1 + m(1 - T_r^{0.5}) \quad (18)$$

$$m = 0.379642 + 1.485030\omega - 0.164423\omega^2 + 0.016666\omega^3 \quad (19)$$

$$b = 0.077796 \frac{RT_c}{P_c} \quad (20)$$

In terms of the compressibility factor (Z), the Peng-Robinson EOS may be written:

$$Z^3 - (1 - B)Z^2 + (A - 2B - 3B^2)Z - (AB - B^2 - B^3) = 0 \quad (21)$$

$$\text{Where, } A = 0.457235 \left(\frac{P_r}{T_r^2} \right) \alpha \quad (22)$$

$$B = 0.077796 \left(\frac{P_r}{T_r} \right) \quad (23)$$

$$\text{Experimentally, } z = \frac{PV}{RT} \quad (24)$$

The Peng-Robinson EOS is generally superior in predicting the liquid densities of many materials, especially non polar ones.

Results and Discussions

In Table 1, the fatty acids whose critical properties and acentric factors were predicted by the Group Contribution methods are presented. The different critical properties and acentric factors of fatty acids calculated using the Group Contribution methods are shown in Table 2. The values of the critical properties of fatty acids predicted by the different Group Contribution methods show a significant similarity. As the number of carbon atoms increases, critical temperature and acentric factor increases monotonically. The reverse is the case for critical pressure. Table 3 is a comparison of the predicted P-V-T data of some fatty acids from the Peng-Robinson EOS with experimental values.

Table 1: Molecular Formular and Some Properties of the Fatty Acids

Common Name	IUPAC Name	Molecular Formula	No. of Carbon Atoms	T_b /(K)	M/(kgmol ⁻¹)
Caproic acid	Hexanoic acid	CH ₃ (CH ₂) ₄ CO ₂ H	6	478.800	0.116
Caprylic acid	Octanoic acid	CH ₃ (CH ₂) ₆ CO ₂ H	8	512.700	0.144
Capric acid	Decanoic acid	CH ₃ (CH ₂) ₈ CO ₂ H	10	543.600	0.172
Lauric acid	Dodecanoic acid	CH ₃ (CH ₂) ₁₀ CO ₂ H	12	571.900	0.200
Myristic acid	Tetradecanoic acid	CH ₃ (CH ₂) ₁₂ CO ₂ H	14	582.000	0.228
Palmitic acid	Hexadecanoic acid	CH ₃ (CH ₂) ₁₄ CO ₂ H	16	605.600	0.256
Stearic acid	Octadecanoic acid	CH ₃ (CH ₂) ₁₆ CO ₂ H	18	628.200	0.284
Oleic acid	Cis-heptadec-8-ene-1-carboxylic acid	CH ₃ (CH ₂) ₇ CH=CH(CH ₂) ₇ CO ₂ H	18	626.817	0.250
Linoleic acid	Heptadeca-8,11-diene-1-carboxylic acid	CH ₃ (CH ₂) ₄ CH=CHCH ₂ CH ₂ CH=CH(CH ₂) ₇ CO ₂ H	18	626.810	0.248
Linolenic acid	Heptadeca-8,11,14-triene-1-carboxylic acid	CH ₂ CH ₂ CH=CHCH ₂ CH=CHCH ₂ CH=CH(CH ₂) ₇ CO ₂ H	18	626.804	0.246

Table 2: Different Critical Properties and Acentric Factors Calculated using the Group Contribution Methods

Fatty Acids	Ambrose	Lydersen			Constantnou and Gani		
	$T_c/(K)$	$T_c/(K)$	$P_c(\text{bar})$	ω	$T_c/(K)$	$P_c(\text{bar})$	ω
Caproic acid	669.557	667.061	32.996	0.655	665.447	34.452	0.683
Caprylic acid	696.727	691.553	26.550	0.750	695.000	27.675	0.760
Capric acid	721.131	713.597	22.208	0.846	720.403	22.785	0.822
Lauric acid	743.230	734.170	19.087	0.935	742.676	19.142	0.884
Myristic acid	743.040	733.945	16.736	1.009	762.508	16.356	0.708
Palmitic acid	761.281	753.445	14.900	1.060	780.381	14.177	0.748
Stearic acid	778.992	774.241	13.427	1.080	796.648	12.440	0.795
Oleic acid	790.051	773.124	12.124	0.990	796.337 \pm 1.167	12.424 \pm 0.260	0.732
Linoleic acid	805.185	773.733	12.340	0.995	796.018 \pm 2.339	12.417 \pm 0.519	0.732
Linolenic acid	823.418	774.376	12.563	1.000	796.648 \pm 3.873	12.418 \pm 0.777	0.731

Table 3: Comparison of Predicted P – V – T Data of some Fatty Acids from the Peng – Robinson EOS with Experimental Values.

Substance	$T/(K)$	$P_c(\text{bar})$	$V/(\text{Lmol}^{-1})$	$Z_{\text{expt.}}$	$Z_{\text{cal.}}$	$100_{\Delta Z/Z_{\text{expt.}}}$
Caprylic acid	343.15	10.0	0.1654	0.058	0.131	-125.86
		30.0	0.1651	0.174	0.144	17.24
		50.0	0.1648	0.289	0.239	17.30
		75.0	0.1644	0.432	0.356	17.59
		90.0	0.1642	0.518	0.463	10.62
Capric acid	353.15	10.0	0.2013	0.069	0.154	-12.32
		30.0	0.2009	0.205	0.173	15.61
		50.0	0.2005	0.341	0.285	16.42
		75.0	0.2000	0.511	0.423	17.22
		90.0	0.1997	0.612	0.514	16.01
Lauric acid	358.15	10.0	0.2365	0.079	0.069	12.66
		30.0	0.2361	0.238	0.203	14.71
		50.0	0.2356	0.396	0.335	15.40
		75.0	0.2351	0.592	0.711	-20.10
		90.0	0.2348	0.710	0.782	-10.14
Myristic acid	363.15	10.0	0.2721	0.090	0.082	8.89
		30.0	0.2716	0.270	0.241	10.74
		50.0	0.2711	0.449	0.397	11.58
		75.0	0.2704	0.672	0.588	12.50
		90.0	0.2701	0.805	0.699	13.17
Palmitic acid	368.15	10.0	0.3080	0.101	0.093	7.92
		30.0	0.3074	0.301	0.275	8.64
		50.0	0.3068	0.501	0.452	9.78
		75.0	0.3061	0.750	0.853	13.73
		90.0	0.3057	0.899	0.895	0.44

A comparative plot of the experimental compressibility factors and the calculated compressibility factors of some fatty acids can be observed from Figures 1-5. There is a good agreement between experimental and predicted compressibility factors. The average percentage error ($100\Delta Z/Z_{\text{expt.}}$) in the estimation of the critical properties of Palmitic acid is less than 10%. For Cayrylic, Capric, Lauric and Myristic acids, it is less than 16%.

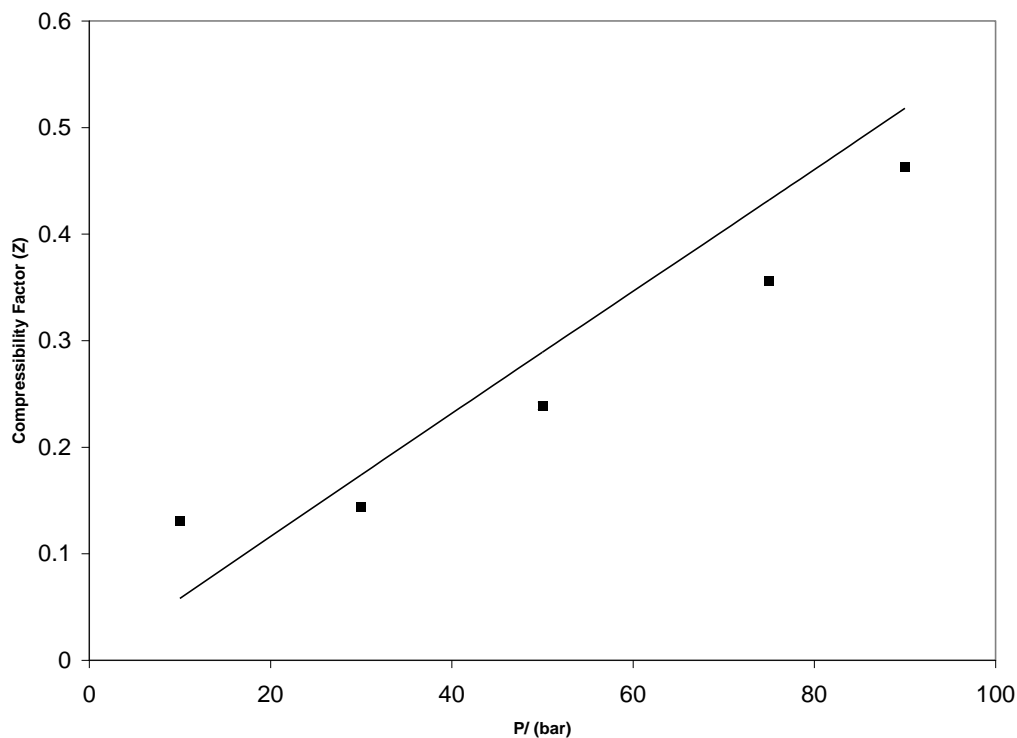


Fig: 1 Comparison of experimental compressibility factors of caprylic acid with calculated values at 343.15K

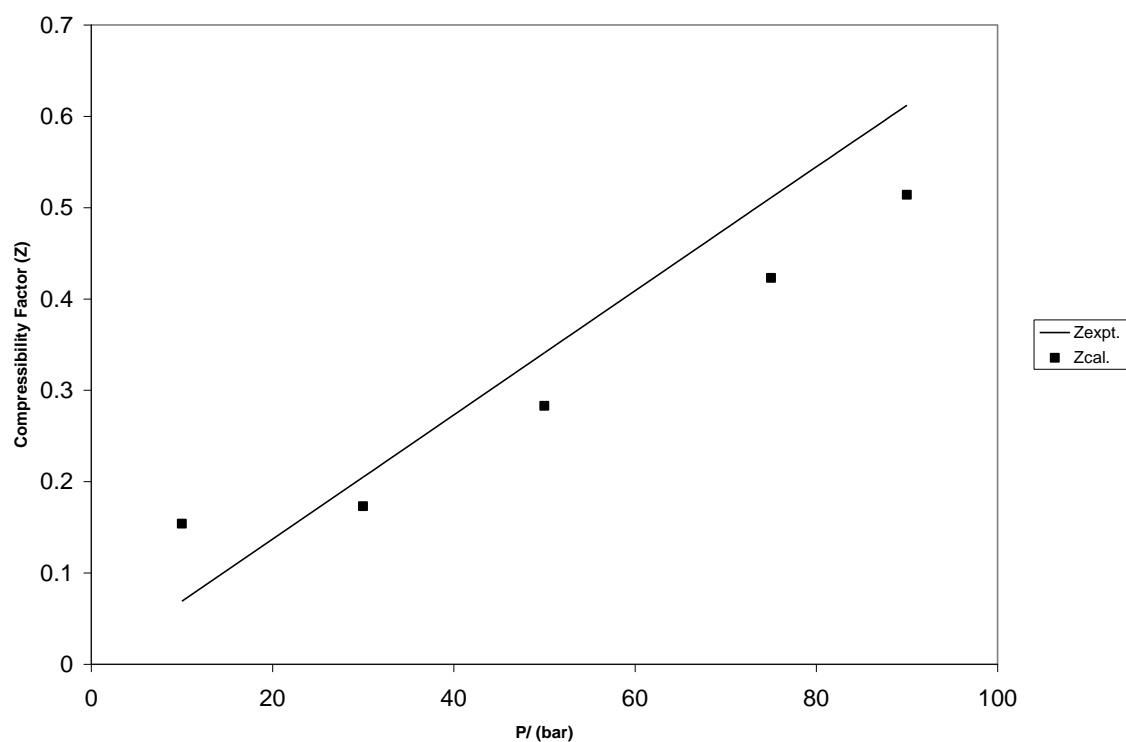


Fig: 2

Comparison of experimental compressibility factors of capric acid with calculated values at 353.15K

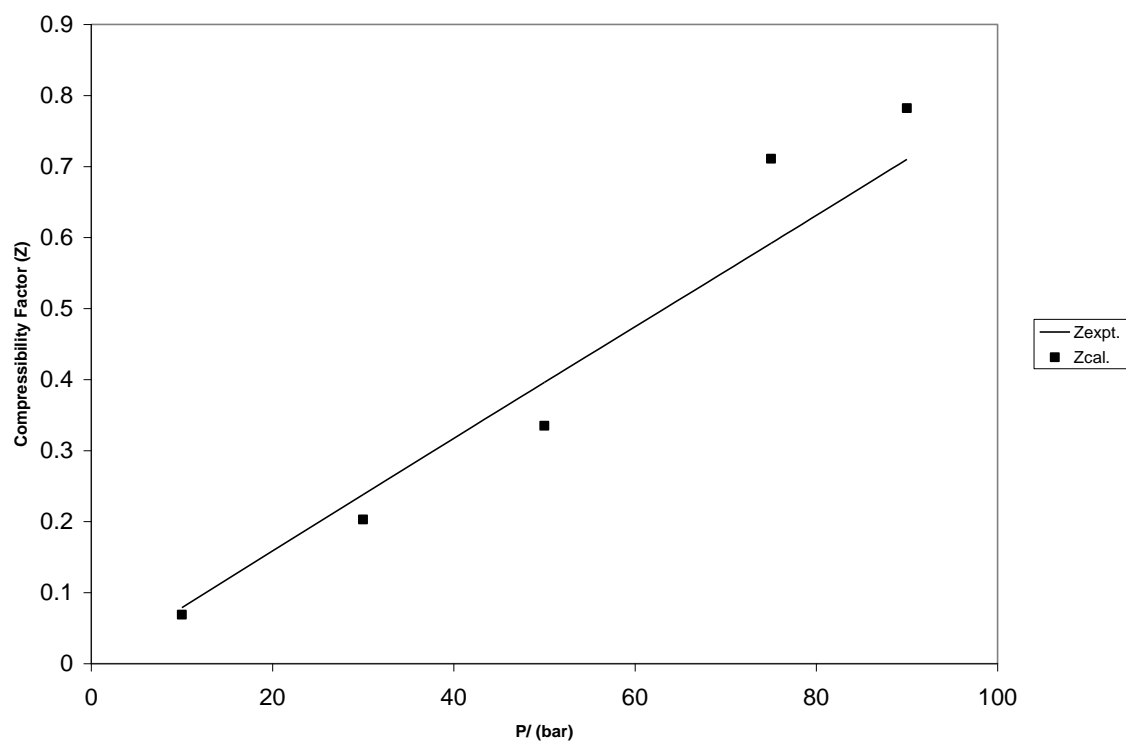


Fig: 3

Comparison of experimental compressibility factors of lauric acid with calculated values at 358.15K

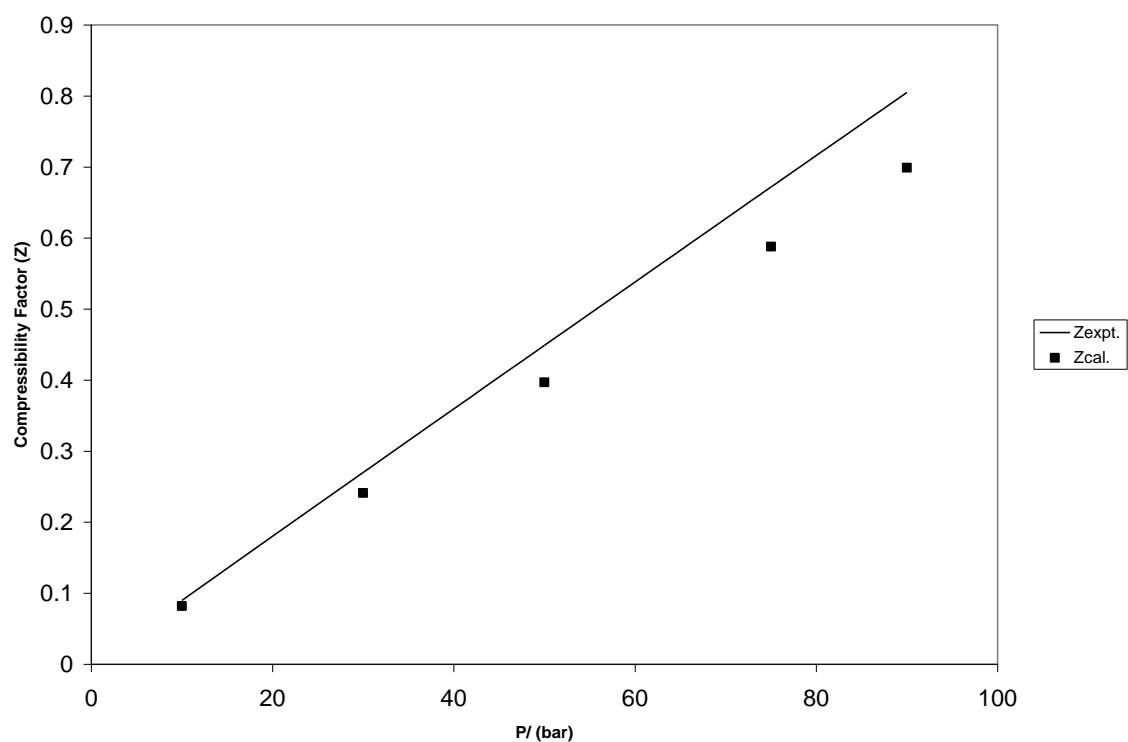


Fig: 4

Comparison of experimental compressibility factors of myristic acid with calculated values at 363.15K

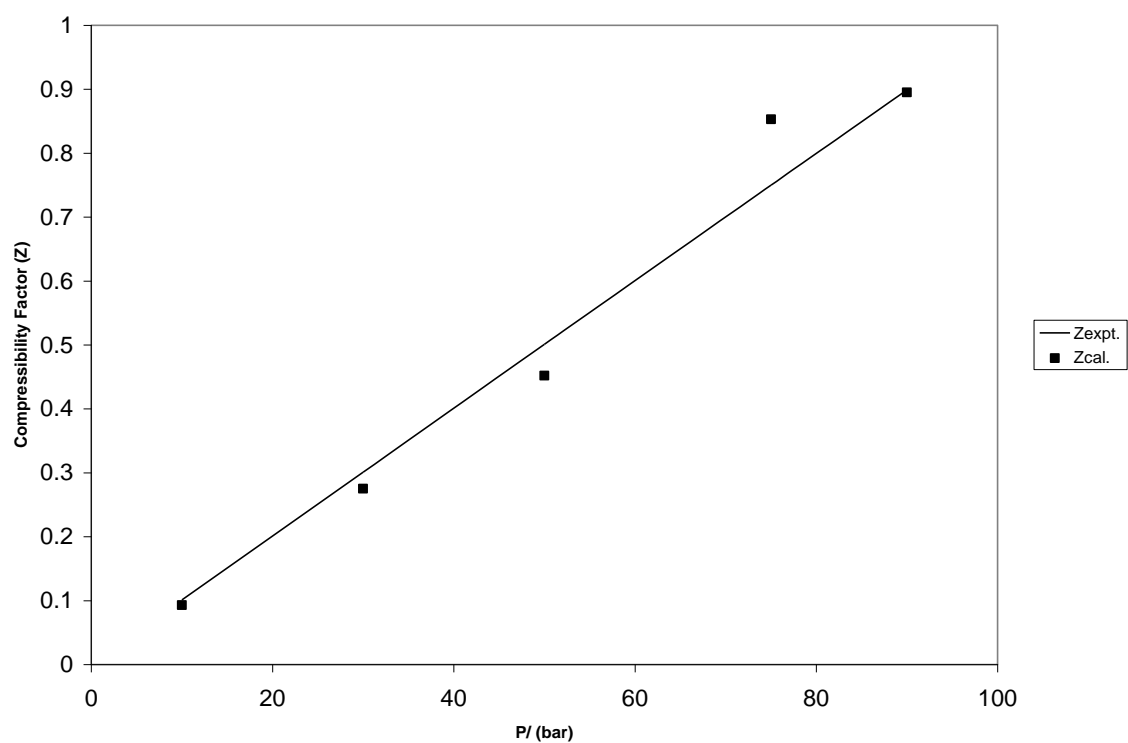


Fig: 5 Comparison of experimental compressibility factors of palmitic acid with calculated values at 368.15K

Conclusion

Three methods for the estimation of critical properties and one method for the estimation of acentric factor of fatty acids were considered and tested for their suitability, with the Peng – Robinson EOS for liquid density calculations. The primary interest was to establish the best methods for extrapolation to fatty acids for which experimental critical data are not available. It has been established that the three methods gave similar values for the critical properties of fatty acids. However for general applicability, the method of Constantinou and Gani [9] is a better choice. The latter method is the only Group Contribution approach not requiring the normal boiling temperature for the estimation of critical temperature. Furthermore, it is probably the best method available today for extrapolations.

This is the first time a research of this nature, which is directly applicable to the palm oil industry is being carried out. It is hoped that these findings will have a major impact in the processing techniques for the separation and purification of palm oil.

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A STUDY OF THE CONCENTRATION OF SEVEN HEAVY METALS IN THE LEAVES OF TWO SPECIES OF PLANT (*Elaeis guineensis* and *Prunus dulcis*) IN STEEL PRODUCTION AREAS OF OVWIAN-ALADJA, DELTA STATE, NIGERIA

S. O. Akporido

Department of Chemistry, Delta State University, Abraka

e-mail: samaccess2006@yahoo.com

Abstract

The concentration of seven heavy metals were determined in the leave samples of oilpalm (*Elaeisis guineensis* and *Prunus dulcis*) for 2 dry and 2 rainy seasons at seven sampling stations in the steel production area of Ovwian-Aladja in Delta state , Nigeria. The average concentration of heavy metals obtained for oilpam leaves are copper (49 ± 18 mg/kg), lead (114 ± 91 mg/kg), nickel (21.9 ± 5.6 mg/Kg), cadmium (6.8 ± 2.0 mg/kg), zinc (250 ± 120 mg/kg), iron (1020 ± 240 mg/kg) and manganese (430 ± 220 mg/kg). The average concentrations of heavy metals in almond leaves (*Prunus dulcis*) are Cu (212 ± 83 mg/L), Pb (190 ± 120 mg/kg), Ni (24.9 ± 8.0 mg/kg), Cd (8.3 ± 3.1 mg/kg), Zn (680 ± 440 mg/kg), Fe (1230 ± 590 mg/kg) and Mn (1000 ± 640 mg/kg). Comparison of the metal concentration of the study with the control area showed that the concentration of all metals in the study area are significantly higher than the concentration of heavy metals in the control area.

Keywords: *Elaeisis guineensis*; *Prunus dulcis*; steel production; heavy metals; copper; lead; nickel; cadmim; zinc; iron; manganese; study area; control area.

INTRODUCTION

Certain activities contribute high proportions of heavy metals into any environment. Among these are the mining and smelting of metals, steel production, atmospheric fall outs brought about by exhaust of motor vehicles (e.g. use of leaded gasoline), combustion of fossil fuels, disposal of urban and industrial waste and waste from use of inorganic fertilizers in agriculture (Forstner and Wittman, 1983). Mining and smelting of metal contribute the highest proportion of heavy metal pollutants to any environment. Effects of individual heavy metals on man, animals and are well documented by Olivera (1994), Ayleit (1979), Nriagu and Pocyna (1988), Langstren (1990), Bryan (1971), Forstner and Wittman (1983) and Sharma and Agrawal (2000).

Work on the determination of heavy metals in plant leaves, plant roots and plant have been carried out by Yusuf et al (2003), Gbamka and Friday (2007) in Nigeria and others, Levy et al (1991), Dentron *et al.* (1980) in other parts of the world. Yusuf et al (2003) found concentrated ranges of Cu ($25.1 - 56.8$ m/kg) Ni ($1.33 - 2.06$ m/kg) and Cd ($1.13 - 1.67$) m/kg) in *Talium triangulare*, *Calisia trigna* and *Ceralus oliterus* plants in industrial areas of Lagos. Levy *et al* (1992) found concentrations of Cu (11.2 mg/kg), Pb (52.0 mg/kg) Cd (7.27 mg/kg) and Zn (517 mg/kg) in *Achillia lanulosa* plants in the mining areas of Arkansas River (Leadville Colorado), USA. Steel production have been taking place in Ovwian – Aladja areas of Delta State, Nigeria in later part of the 1980's. The extent to which this activity have impacted on the area have not been well studied. The objectives of this study is to assess the impact of the activity of steel production on the area, especially the vegetation by the determination of selected heavy metals in two species of plant that is the leaves of oil palm tree (*Elaeis guineensis*) and the almond tree (*Prunus dulcis*).

MATERIALS AND METHOD

Description of study area:

Area under study is given in Fig. I (Map of study area), sampling stations and designated A, B, C, D, E, F and G with sampling station A very near to the Delta Steel Company (DSC) and sampling station G is farthest from DSC along the Udu River (section of Warri River) which also coincides with the Local Government Area boundary between Warri and Udu LGA. Each sampling is separated from the next successive sampling station by 1 km distance.

Design of Study: Samples were collected twice every season for two years (i.e. dry and rainy seasons) starting from January 2007 to September 2008. The heavy metals determined in the leaves of the two plant species are Cu, Pb, Ni, Cd, Zn, Fe and Mn.

Samples were also collected from two points around Ovwuvwe River at Abraka Inland in Ethiope East which serves as control samples.

Sample Collection/Preparation

Fresh shoots of leaves were collected using sharp plastic materials and these were transferred to the laboratory in polythene bags inside coolers containing iced blocks. In the laboratory, they were dried in the oven at 60 °C for 24 hrs.

Analytical Procedures

Digestion of Leave Samples:

To 0.5g of dried and well ground leave samples in a kjedahl flask was added 5 ml of concentrated nitric acid, 1 ml of 60% per chloric acid and finally 0.5 ml of concentrated sulphuric acid, details as described by Allens (1989).

AAS Analysis of Digest Solution: The digest solution was aspirated into an AAS spectrophotometer that has already been calibrated with standard solutions of the metals being determined. Details is as described in Allens (1989).

Quality Assurance Programme :

Among the quality control measures taken are the following:

- i. Good representative sampling were carried out, cleanliness of laboratory and equipment used were observed.
- ii. Blank determinations were carried out.
- iii. Percentage recoveries of each metal from samples of both oil palm tree and almond tree leaves were carried out. A measured amount of well ground and leaves sample was spiked with a known concentration of metal (standard). This was allowed to get dried in an oven. This was homogenated with a glass rod. 0.5g of this spiked and homogenated samples was analyzed using the same procedure as described above for the samples. The concentration obtained in this case is the concentration of the re-analysis of the same sample after it has been spiked with the metal standards. Results of percentage recoveries of the metals from oil palm leave samples are 93.4%, 91.5%, 97.2%, 92.8%, 101%, 99%, and 10.3 for Cu, Pb, Ni, Cd, Zn, Fe and Mn respectively. The percentage recoveries of metals from almond tree leaves are 94%, 96.7%, 93.3%, 91.8%, 101%, 95% and 99% for Cu, Pb, Ni, Cd, Zn, Fe and Mn respectively. The method of analysis is thus a good one.

RESULTS AND DISCUSSION

The concentrations of the metals in the four seasons (Table I and II) are not statistically significantly different from one another (ANOVA – SINGLE FACTOR)

The concentrations of the seven heavy metals in the leaves of the two plants in the study area were also compared to their concentrations in the control area (control area is area around Ovwuvwe Stream in Abraka Inland, Ethiope East LGA). Table III and Table IV shows a comparison

of concentrations of heavy metals in oil palm leaves and almond leaves of study areas respectively with the concentration of heavy metals in the control area.

With the exception Cd in almonds, the concentrations of oil heavy metals in leaves in both oil palm and almonds in the sandy area are statistically significantly higher than their concentrations in the control area when compared using t - test (two sample – assuming equal variance). This shows that the study area is polluted with respect to these heavy metals relatively to the control area. The concentrations of heavy metals in the study area were also correlated with each other. Table V shows a Pearson (2-tailed) correlation of heavy metals in leaves of oil palm.

Table V shows that the seven heavy metals correlates very strongly with each other, the coefficient of correlation in each case is significant at 99% confidence level. This indicates that the heavy metals have identical sources. The source of the metal is most likely the iron and steel industry located in the area. Since there are no other notable industry or activity that can result in the release of all these heavy metals to the environment at such concentration.

Table VI shows a comparison of results of determination of heavy metals in plant from studies elsewhere with the results obtained for the present study. It shows that the results are comparable with results obtained for other plants species elsewhere. The average concentration for Cu in *Elaeis guineensis* (49 ± 18 mg/kg) and *prunus dulcis* (212 ± 83 mg/kg) in the study area are comparable to the range of concentrations obtained for *Talium triangulare*, *Celisia trigna* and *Carclus oliterus* (25.1 – 56.8 mg/kg) from the industrial area of Lagos, *Agropyron spp* in mining area of Arkansas river (8.10 mg/kg), *Heliphica spp* in sea Port of Cape York, Australia (9.00 mg/kg) and *Achilla lanulosa* plant of the mining area of Arkansas River in Leadville, Colorado (11.2 mg/kg). The average concentrations of Cu in the two species in the study area are higher than the concentration ranges normally encountered in plants (2.50 – 25.0 mg/kg) (Allens, 1989). This shows an enhancement in the concentration of Cu in the study area which also means Cu has been anthropogenically inputed into the environment in this area i.e. the area is polluted with respect to Cu. The average concentration of Pb in *E. guineensis* (114 ± 91 mg/kg) in the study area are comparable to that obtained for *Achillia lanulosa* in Arkansas River mining area, USA (52.0 mg/kg) *Iris missouriensis* of the same area (23.4 mg/kg) but higher than those for *Hibiscus esculenta* (root) in Niger Delta oil prospecting area (0.50 ± 0.03 mg/kg), *H. esculenta* (stem) (0.41 ± 0.02 mg/kg) *H. esculenta* (leaves) (0.37 ± 0.01 mg/kg) and *H. esculenta* (fruit) (0.22 ± 0.03 mg/kg) of the same area and in the same study. The average concentrations of Pb in the two species of the study are also very much higher than the concentration ranges normally encountered in plant (0.05 – 3.00 mg/kg) thus showing that the area is polluted with respect to this metal. The average concentration of Ni in *E. guineensis* (21.9 ± 5.6 mg/kg) and *P. dulcis* (24.9 ± 8.0 mg/kg) are comparable with the range of concentrations found in three species: *Telim triangulare*, *Celisa trigna* and *Carclus oliterus* (1.33 – 2.06 mg/kg) and *Helophica spp* (1.70 mg/kg) of the mining area around Arkansas River, Leadville Colorado, USA. The average concentrations of Ni in both species also exceeds the concentration ranges normally encountered in plants (0.50 – 5.00 mg/kg) showing that the area may be polluted with respect to Ni. The average concentrations of Cd in *E. guineensis* (6.8 ± 2.0 mg/kg) and *P. dulcis* (8.3 ± 3.1 mg/kg) are comparable to concentration ranges obtained for three species: *Talim triangulare*, *Celisia trigna* and *Carclus oliterus* (1.13 – 1.67 mg/kg), *Achillia lanulosa* (7.27 mg/kg) and *Iris missouriensis* (21.0 mg/kg). The average concentration of Cd in the two species of plants in the study area also exceed the concentration range normally encountered in plant materials (0.01 – 0.30 mg/kg). This indicates that the area may be polluted with respect to Cd.

The average concentration of Fe in *E. guineensis* (1020 ± 240 mg/kg) and *P. dulcis* (1230 ± 590 mg/kg) by far exceeds the normal concentration range of Fe encountered in plant materials (40.0 – 500 mg/kg) which indicates that the area is polluted with respect to Fe. The average concentrations of Mn in *E. guineensis* (430 ± 220 mg/kg) and *P. dulcis* (1000 ± 640 mg/kg) appears to fall within the concentration ranges normally encountered in plant materials (50 – 1000 mg/kg) concentration of Mn in *P. dulcis* (1000 ± 640 mg/kg) appears to be in the high side of this range, thus indicating the increasing input of Mn to the environment.

CONCLUSION

Concentration of heavy metals were determined in the leaves of oil palm tree (*E. guineensis*) and almond tree (*P. dulcis*). The concentrations found in the study area were statistically and significantly higher than in the control area for each of the heavy metals. The average concentration of the heavy metals in the leaves of these two plant species were comparable to results obtained elsewhere i.e. areas which could be said to be polluted. The average concentration of the seven heavy metals in the leaves of the two plants were also higher than ranges of concentrations of corresponding heavy metals normally encountered in plant material. These also go to show that the area is polluted with respect to the seven heavy metals. Anthropogenic input of the metals has also been established.

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Table I: Average concentration of the metals in leaves of oil palm (*Elaeis guineensis*) in each season

Heavy Metals	First dry season	First rainy season	Second dry season	Second rainy season
Cu (mg/kg)	46 ± 18	47 ± 20	46 ± 18	46 ± 20
Pb (mg/kg)	142 ± 110	145 ± 110	141 ± 110	145 ± 110
Cd (mg/kg)	6.4 ± 2.6	6.5 ± 2.6	6.5 ± 2.6	6.5 ± 2.5
Ni (mg/kg)	21.0 ± 4.2	21.0 ± 4.2	20.8 ± 4.1	21.5 ± 4.4
Fe (mg/kg)	1050 ± 190	1060 ± 200	1040 ± 190	1060 ± 200
Mn (mg/kg)	350 ± 180	356 ± 180	356 ± 190	357 ± 180
Zn (mg/kg)	230 ± 120	236 ± 120	232 ± 120	233 ± 120

Table II: Average concentration of the heavy metals in leaves of almond tree (*Prunus dulcis*) in each of the seasons.

Heavy Metals	First dry season	First rainy season	Second dry season	Second rainy season
Cu (mg/kg)	211 ± 68	216 ± 200	219 ± 71	229 ± 64
Pb (mg/kg)	184 ± 120	188 ± 130	187 ± 120	182 ± 130
Cd (mg/kg)	7.9 ± 3.3	8.1 ± 2.9	8.0 ± 2.7	8.5 ± 3.0
Zn (mg/kg)	611 ± 420	623 ± 430	633 ± 420	643 ± 430
Ni (mg/kg)	24.9 ± 6.7	26.0 ± 6.7	25.1 ± 7.6	27.4 ± 6.6
Fe (mg/kg)	1240 ± 570	1250 ± 570	1280 ± 570	1300 ± 590
Mn (mg/kg)	910 ± 720	931 ± 740	921 ± 690	921 ± 740

Table III: Concentrations of heavy metals in oil palm leaves in the study and control areas.

Heavy metals	Study area	Control area
Cu (mg/kg)	49 ± 18	16.1 ± 3.0
Pb (mg/kg)	114 ± 91	11.6 ± 1.7
Ni (mg/kg)	21.9 ± 5.6	7.2 ± 1.1
Cd (mg/kg)	6.8 ± 2.0	1.8 ± 0.1
Zn (mg/kg)	250 ± 120	20.3 ± 3.5
Fe (mg/kg)	1020 ± 240	26.8 ± 4.6
Mn (mg/kg)	430 ± 220	21.4 ± 2.0

Table IV: Concentrations of heavy metals in oil palm leaves in the study and control areas

Heavy metals	Study area	Control area
Cu (mg/kg)	212 ± 83	15.2 ± 1.2
Pb (mg/kg)	190 ± 120	13.6 ± 0.5
Ni (mg/kg)	24.9 ± 8.0	14.6 ± 0.7
Cd (mg/kg)	8.3 ± 3.1	6.20 ± 0.60
Zn (mg/kg)	680 ± 440	21.1 ± 1.4
Fe (mg/kg)	1230 ± 590	23.1 ± 4.8
Mn (mg/kg)	1000 ± 640	17.0 ± 2.5

Table V: Pearson (2-tailed) correlation of heavy metals in leaves of oil palm (*Elaeis guineensis*)

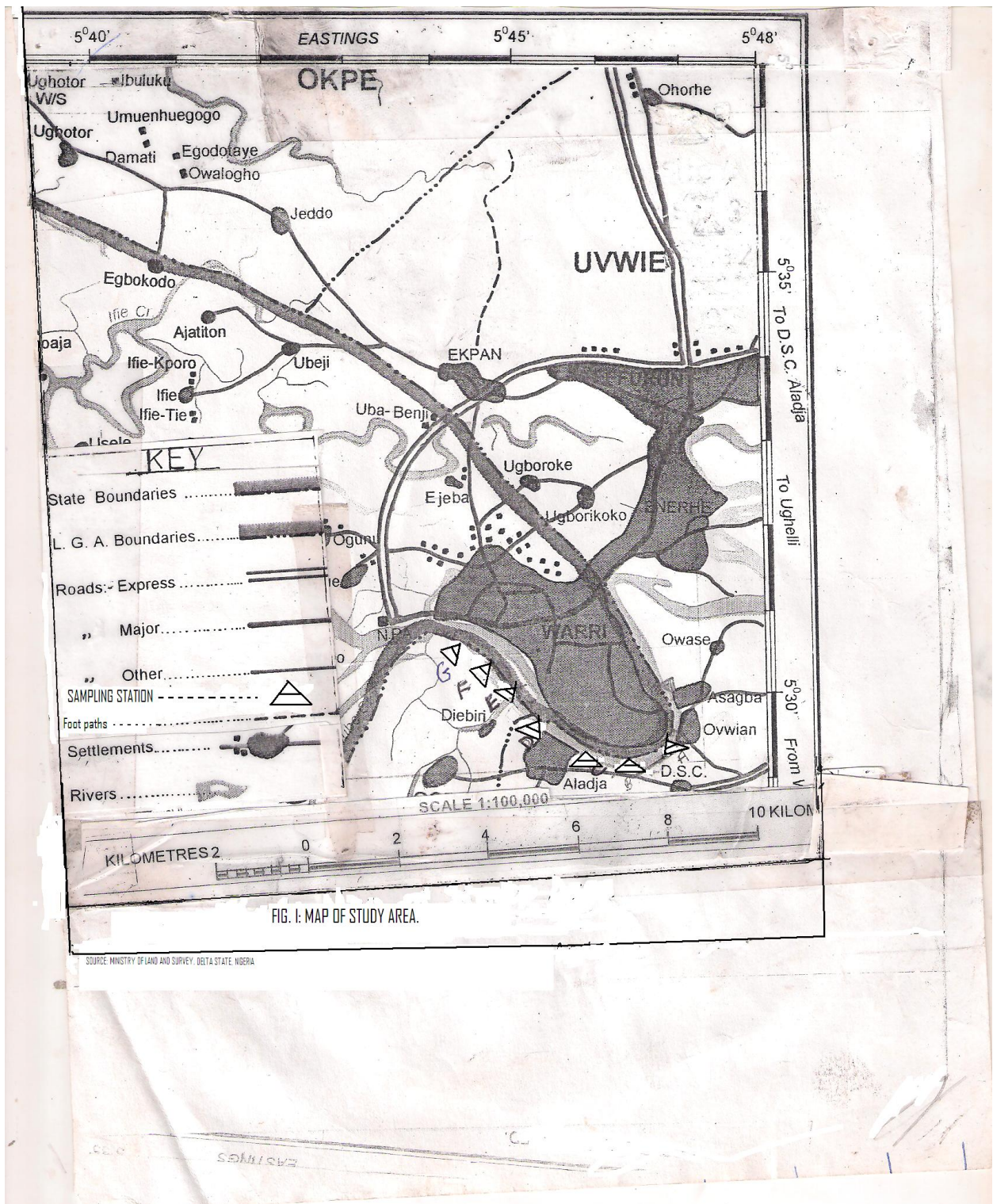
	Cu	Pb	Ni	Cd	Zn	Fe	Mn
Cu							
Pb	0.587**						
Ni	0.896**	0.685**					
Cd	0.84**	0.602**	0.780**				
Zn	0.965**	0.649**	0.847**	0.881**			
Fe	0.884**	0.621**	0.827**	0.927**	0.902**		
Mn	0.828**	0.432**	0.687**	0.824**	0.799**	0.850**	

** Coefficient of correlation significant ($\alpha = 0.01$)

* Coefficient of correlation significant ($\alpha = 0.05$)

Table VI: Results of determinants of heavy metals in plant from studies elsewhere compared with result for perfect study and re-concentration ranges normally encountered in plants.

Country	Location	Major activities in area	Species of plant	Cu (mg/kg)	Pb (mg/kg)	Ni (mg/kg)	Cd (mg/kg)	Zn (mg/kg)	Fe (mg/kg)	Mn (mg/kg)	References
Nigeria	Lagos	Industrial	(i) Talim triangulare (ii) Celisa trigina (iii) Carclus olitarus	25.1 – 56.8		1.33 – 2.06	1.13 -1.67				Yusuf et al., 2003
Nigeria	Niger Delta	Oil prospecting	(i) Hibiscus esculenta (root)	0.20 ± 0.03	0.50 ± 0.03	0.16 ± 0.04	-	1.83 ± 0.05			Gbaruka and Friday, 2007
			(ii) Hibiscus esculenta (stem)	0.16 ± 0.01	0.41 ± 0.02	0.41 ± 0.01		1.10 ± 0.00			
			(iii) Hibiscus esculenta (leaf)	0.16 ± 0.01	0.37 ± 0.01	0.13 ± 0.01		0.88 ± 0.02			
			(iii) Hibiscus esculenta (fruit)	0.12 ± 0.030	0.22 ± 0.03	0.08 ± 0.01		0.65 ± 0.03			
USA	Arkansas River, Leadville Colorado	Mining	Agropyron spp	8.10	1.06		0.27	61.0			Levy et al
Austria	Cape York	Sea Port	Helophila spp	9.00	1.00	1.70	0.50	67.0			Denton et al, 1980
USA	Arkansas River, Leadville Colorado	Mining	Achilia Lanulosa	11.2	52.0		7.27	517			Levy et al
USA	Arkansas River, Leadville Colorado	Mining	Iris missouriensis	4.70	23.4		21.0	40.3			Levy et al
Nigeria	Udu River (Aladja/Ovwian)	Steel manufacturing	Eleais guineensis	49 ± 180	114 ± 91	21.9 ± 5.6	6.8 ± 2.0	250 ± 120	1020 ± 240	430 ± 220	Present study
Nigeria	Udu River (Aladja/Ovwian)	Steel manufacturing	Prumis dulcis	212 ± 83	190 ± 120	241 ± 8.0	8.3 ± 3.1	680 ± 440	1230 ± 590	1000 ± 640	Present study
Concentration ranges normally encountered in plant				2.50 – 25.0	0.05 – 3.00	0.50 – 5.00	0.01 – 0.30	15.0 – 100	40 – 500	50 – 1000	Allens 1989



AN ESTIMATION OF THE pH AND HEAVY METALS IN THE COMBINED BREWERY EFFLUENTS OF BENDEL BREWERY LTD. AND GUINNESS NIGERIA PLC AND WATER OF THE RECEIVING IKPOBA RIVER

S. O. Akporido

DEPARTMENT OF CHEMISTRY, DELTA STATE UNIVERSITY, ABRAKA

Abstract

A study of the quality of the combined effluents from the twin breweries of Bendel Brewery Ltd. And Guinness Nigeria Plc and water of the receiving Ikpoba River was carried out. Effluent samples collected from the combined effluent conduit and water samples from the receiving river were analyzed for the parameters of temperature, pH, Lead, Copper, Cadmium and Chromium. Results for Pb ($419 \pm 360 \mu\text{g/L}$) and Cd ($30 \pm 28 \mu\text{g/L}$) in effluent exceeds two national effluent guidelines. Result for Pb ($202 \pm 80 \mu\text{g/L}$), Cr ($67 \pm 64 \mu\text{g/L}$) and Cd ($21 \pm 23 \mu\text{g/L}$) in the receiving water exceed several international and national drinking water guidelines. The average value for Pb in water exceeds guidelines for irrigation and livestock water, Cu ($37 \pm 11 \mu\text{g/L}$) and Cd ($21 \pm 23 \mu\text{g/L}$) also exceed irrigation guidelines.

Keywords: Combined Effluent Conduit, Guinness Nigeria Plc, Bendel Brewery Ltd., Copper, Cadmium, Chromium, pH, Irrigation guidelines, Effluent Guidelines, Drinking Water Guidelines.

INTRODUCTION

Industrialization in a country almost always is accompanied with problems of environmental degradation. Establishment of breweries is a good contribution to the industrial growth of a nation. Effluents from brewery industries may contribute negatively to efforts to have an unpolluted environment. Untreated brewery effluents contain suspended solids ($10 - 60 \text{ mg/kg}$), biochemical oxygen demand (BOD_5) ($1000 - 1500 \text{ mg/kg}$), chemical oxygen demand (COD) ($800 - 3000 \text{ mg/L}$) (World Bank, 1997). Brewery effluents is also known to have genotoxic effect in *Clarias lazira* (Odeigah and Osanyipeju, 1995). Wastewater from industries such as the breweries also brings about variation in bulk properties of anaerobic granules (Batstone and Keller, 2001). Analysis of brewery effluents i.e xanthohunol and related phenylflavanoids in hops and beer by liquid chromatography-tandem mass spectrometry has been carried out (Keyser *et al.*, 1999).

MATERIALS AND METHOD

Description of study area:

The two breweries are located in South of Benin City in the Southern part of Nigeria. The Ikpoba River also lies to the South of Benin City. The main effluent drain from the two breweries converge at a point before continuing to the Ikpoba River. The description is illustrated by Fig. 1 (Map of the Study area (A Section of Benin City)).

Design of Study: Effluent Samples were taken from point of combination of the two effluents (PCE), at a 100m from that point (200m PCE). Water samples from the receiving river were collected at five points: Point of entry of effluents into the river (PEE), 500m downstream from point of entry of effluents into the river (500m DSPEE), at 1.5 km downstream from point of Entry of Effluents into the river (1km DSPEE), 500m upstream from point of entry of effluents into the river (500m USPEE) and at 1km North of point of entry of effluents into the river (1km USPEE). Samples were taken twice in every seasons (dry and rainy seasons) for three years - from July 1998 to April 2000. The parameters sampled for and analyzed are mainly the pH and four heavy metals (Pb, Cd, Cu and Cr).

Sample Collection/Preparation

Effluent samples were collected by the time – composite method: discrete samples were collected every 10 mins for 1 hour 10 mins (seven times). These were composited by mixing. The samples were kept at temperature below 4°C. Water samples were collected by the discrete method. effluent and water samples were preserved as stipulated in APHA – AWWA – WEF (1995).

Analytical Procedures

pH:

pH of effluent and water samples were determined at site with a portable pH meter as stipulated in APHA – AWWA – WEF, (1995).

Heavy Metals:

Effluent and water samples were digested by adding 5cm³ of conc. nitric acid to 500ml of samples in Kjeldahl flask and evaporating to near dryness. 20 ml of distilled water was added and this was filtered through a filter funnel containing a No. 44 Whatman filter paper into a 50ml volumetric flask. This was made up to the 50ml mark by the addition of distilled water. The digest solution was taken for atomic absorption spectrometer measurement. Digest solution was aspirated in AAS that has earlier been calibrated with standard solutions of the four heavy metals (Pb, Cu, Cd and Cr). The standard solution were prepared from commercially obtained stock solutions of the heavy metals. Details of the determination of concentration of heavy metals using AAS is in accordance to that described in manual by Allens (1989).

Quality Assurance Programme

A good sampling programme which ensured that representative samples were taken was put into place. Blanks were determined and subtracted from initial sample concentration to get actual concentrations of each of the four metals. Percentage recovery of each metal studied was carried out by spiking a given volume of water sample with a definite concentration of metal solution and re-analysing the sample. Results of recovery studies shows that metals have percentage recoveries of 93.5%, 95.1%, 90.7% and 101% for Pb, Cu, Cd and Cr respectively. The method used for analysis is thus a good one.

RESULTS AND DISCUSSION

A comparison of the values of pH and heavy metals in the seasons (Table I) shows that the change in concentrations of heavy metals with seasons is not statistically significant (ANOVA – Single factor). The average concentration of lead are highest in the second rainy season (209 ± 140 µg/L); The average concentration of Cu, Cd and Cr are highest in the second dry season.

The average concentration of the four heavy metals in the combined effluent was compared with two national effluent guidelines i.e. FEPA (1991) and DPR (2000) (Table II). The average concentration of Pb in effluent (419 ± 470 µg/L) is higher than the guideline values for FEPA (1991)

(50.0 µg/L) and DPR (2000) (50.0 µg/L). The average concentration of Cd (30 ± 51 µg/L) in effluent far exceeds the FEPA (1991) (10.0 µg/L) effluent guideline. These two situation shows that the effluent (i.e. the combined effluent) did not meet environmentally healthy standards. The effluents(or one of the effluents) have not been properly treated. The effluents thus have potentials to pollute any river receiving the effluents.

The average concentrations of the four heavy metals and average value of pH in the receiving river water were compared to international and national drinking water guidelines (Table III). The average pH of the study area (6.23 ± 0.40) falls lower than the guideline range for USEPA (SDWR) (6.5 – 8.5), SON Maximum Permitted Level (MPL) (6.5 – 8.5) and the Canadian Maximum Acceptable Limits (MAC) (6.5 – 8.5). The average concentration of Pb (202 ± 80 µg/L) in receiving water exceeds guideline values for USEPA maximum contamination level (MCL) (15.0 µg/L), WHO 2006 (10.0 µg/L), SON Maximum Permitted Level (MPL) (10.0 µg/L), Canadian Maximum Acceptable Level (MAL) (50.0 µg/L). Also the average concentration of Cd (21 ± 23 µg/L) exceeds all the guideline value i.e. WHO 2006 (3.00 µg/L), USEPA (5.00 µg/L), SON (3.00 µg/L), Canadian (5.00 µg/L) and EEC (5.00 µg/L). The average concentration of Cr (67 ± 64 µg/L) exceeds guideline value for WHO 2006 (50.0 µg/L), SON (50.0 µg/L), Canadian (50.0 µg/L) and EEC (50.0µg/L). From the foregoing it can be said that the water of the receiving water is not suitable for drinking.

The average concentrations of the parameters were also compared to guideline values for non-drinking water uses (Table IV). From table IV, it can be seen that the average pH range for the study area water (6.23 ± 0.40) is lower than the guideline ranges for Aquatic life rearing water (6.5 – 8.5), irrigation water (6.5 – 8.5), and iron and steel industry water (6.8 – 7.0). The average concentration of Lead in the study area water (202 ± 80 µg/L) exceeds guideline values for Livestock water (100 µg/L) and irrigation water (10.0 µg/L). Also the average concentration of Cd in study area water (21 ± 23 µg/L) exceeds guideline value for irrigation water (10.0 µg/L). The water of study area is therefore not suitable for livestock rearing and for irrigation. It must however be cautioned here that the guidelines used are not in operation in Nigeria and they are only used to assess the quality of the water.

The water of the study area (i.e. from point of entry of the effluent downstream) was compared with water of the control area (i.e. water upstream to the point of entry of effluents into the river) (Table V). The average value of pH is slightly lower in the study area than in the control area. This means water of the study area is more acidic than that of the control area. The average concentrations of the four heavy metals are higher in the study area than in the control area. In the case of Cr the difference in the average concentration in the study area and in the control area is statistically significant when compared using T-test (two sample, assuming unequal variance). This confirms that the study area (i.e. downstream from point of entry of effluent into the river) is to some extent more polluted than the control area (i.e. upstream from point of entry of effluents into the river). Since the only factor that could have been responsible for this is the entry of effluents into the river at the point which it did the combined effluents must have been responsible for the observed pollution of that section of the river (i.e. the study area).

Table VI shows the correlation of the concentrations of the four heavy metals. The correlation coefficient of four pair of heavy metals are significant at 0.01 level (2-tailed). They are Cr and Cd (0.978), Pb and Cu (0.808), Pb and Cd (0.837), and Pb and Cr (0.831). Also the correlation of two pair of metals are significant at 0.05 level (2-tailed). They are Cd and Cu (-0.507) and Cr and Cu (-0.801). This means that all the metals correlate strongly with each other. This also show that the four heavy metals observed in the receiving water have identical source. Since the metals were also observed in the effluents, the sources of the metals should be the effluents.

CONCLUSION

Analysis of pH and four heavy metals (Pb, Cu, Cd and Cr) in Brewery effluents and the receiving waters of Ikpoba river has shown that the concentrations of the metals in effluent exceed national effluent guidelines for the metals. The concentration of the metals in the receiving water also exceed national and international guidelines for drinking water and they also exceed international guidelines for non-drinking water users. The average pH of water fall below. most of the national and international guideline for drinking and non-drinking water. All this goes to show that the water of the receiving river water is polluted and will need further treatment before it can be used for drinking purposes and for some of the non-drinking water uses.

Finally, a comparison of average concentration of heavy metals of study area water (downstream from point of effluent entry into the river) and the control area water (upstream from point of entry of effluent) shows that the average concentration of metals in the study area are higher than those of the control which shows that the entry of effluent at that point must have influenced the difference. Also correlation of the heavy metals indicate that they have identical source. Thus it can be safely concluded that the receiving water of Ikpoba river is polluted and the observed pollution is derived from the combined effluents from the Twin brewery of Bendel Brewery Limited and Guinness Brewery Plc.

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Table I: Comparison of pH Levels and average concentration of heavy metals in the Seasons

Heavy Metals	First rainy season	First dry season	Second rainy season	Second dry Season	Third rainy season	Third dry season
pH	6.26 ± 0.48	6.12 ± 0.39	6.42 ± 0.42	6.07 ± 0.32	6.42±0.42	6.12±0.42
Pb (µg/L)	202 ± 130	202 ± 120	209 ± 140	212 ± 140	199 ± 140	196 ± 140
Cu (µg/L)	37 ± 18	37 ± 22	38 ± 29	39 ± 28	37 ± 29	36 ± 29
Cd (µg/L)	21.0 ± 5.6	21.6 ± 7.4	22 ± 11	23 ± 11	23 ± 11	20 ± 11
Cr (Total) (µg/L)	67 ± 34	67 ± 33	69 ± 31	72 ± 33	66 ± 33	63 ± 28

Table II: Average Concentration of heavy metals in Effluent compared to FEPA (1991) and DPR (2000) Effluent Guidelines

Heavy Metals	Average concn. of heavy metals in effluents	FEPA (1991) Guidelines	DPR (2000) Guidelines
Pb (µg/L)	419 ± 470	50.0	50.0
Cu (µg/L)	55 ± 33	1000	1000
Cd (µg/L)	30 ± 51	10.0	No guideline
Cr (Total) (µg/L)	26.8 ± 5.2	30.0	50.0

FEPA = Federal Environmental Protection Agency

DPR = Department of Petroleum Resources

Table III: Comparison of average concentrations of heavy metals and pH of study area with national and international drinking water guidelines

Heavy Metals	Average values	WHO 2006 guidelines (WHO, 2006)	USEPA 2004 MCL (USEPA, 2004)	Canadian MAL (CCREM, 1987)	Soil 2007 MPL (SON, 2007)	EECMA (Sayre, 1988)
pH	6.23 ± 0.40	No guidelines	6.5 – 8.5*	6.5 – 8.5	6.5 – 8.5	No guideline
Pb (µg/L)	202 ± 80	10.0	15.0	50.0	10.0	50.0
Cu (µg/L)	37 ± 11	2000	1300	1000	1000	No guidelines
Cd (µg/L)	21 ± 23	3.00	5.00	5.00	3.00	5.00
Cr (Total) (µg/L)	67 ± 64	50.0	100	50.0	50.0	50.0

WHO = World Health Organisation

USEPA = United State Environmental Protection Agency

SON = Standard Organisation of Nigeria

CCREM = Canadian Council of Resources and Environment Ministers

EEC = European Economic Commission

MCL = Maximum Contaminant Level

MAL = Maximum acceptable Levels

MPL = Maximum Permitted Level

MAC = Maximum Admissible Concentrations

*USEPA Secondary Drinking Water regulation (SOWR) (non-enforceable)

Table IV: Comparison of concentrations of heavy metals and pH levels of study area with guidelines values for some non-drinking water uses

Heavy Metals	Average values	Guidelines for Aquatic Life (Fresh water) (CSWQCB, 1963) in Leeden (1990)	Guideline for Livestock (Ontario Min. of Environment, 1984)	Guideline for Irrigation water (FAO, 1985)	Guideline for Iron and Steel Industry Water (CCREM, 1989)
pH	6.23 ± 0.40	6.5 – 8.5	6.0 – 8.5	6.5 – 8.5	6.8 – 7.0
Pb (µg/L)	202 ± 80	No guideline	100	10.0	No guideline
Cu (µg/L)	37 ± 11	No guideline	500	200	No guideline
Cd (µg/L)	21 ± 23	No guideline	No guideline	10.0	No guideline
Cr (Total) (µg/L)	67 ± 64	No guideline	1000	No guideline	No guideline

CSWQCB = California State water Quality Control Board

FAO = Food and Agricultural Organisation

CCREM = Canadian Council of Resources and Environment Ministers

Table V: Comparison Values of pH and Concentration of heavy metal of water of study area with that of control area.

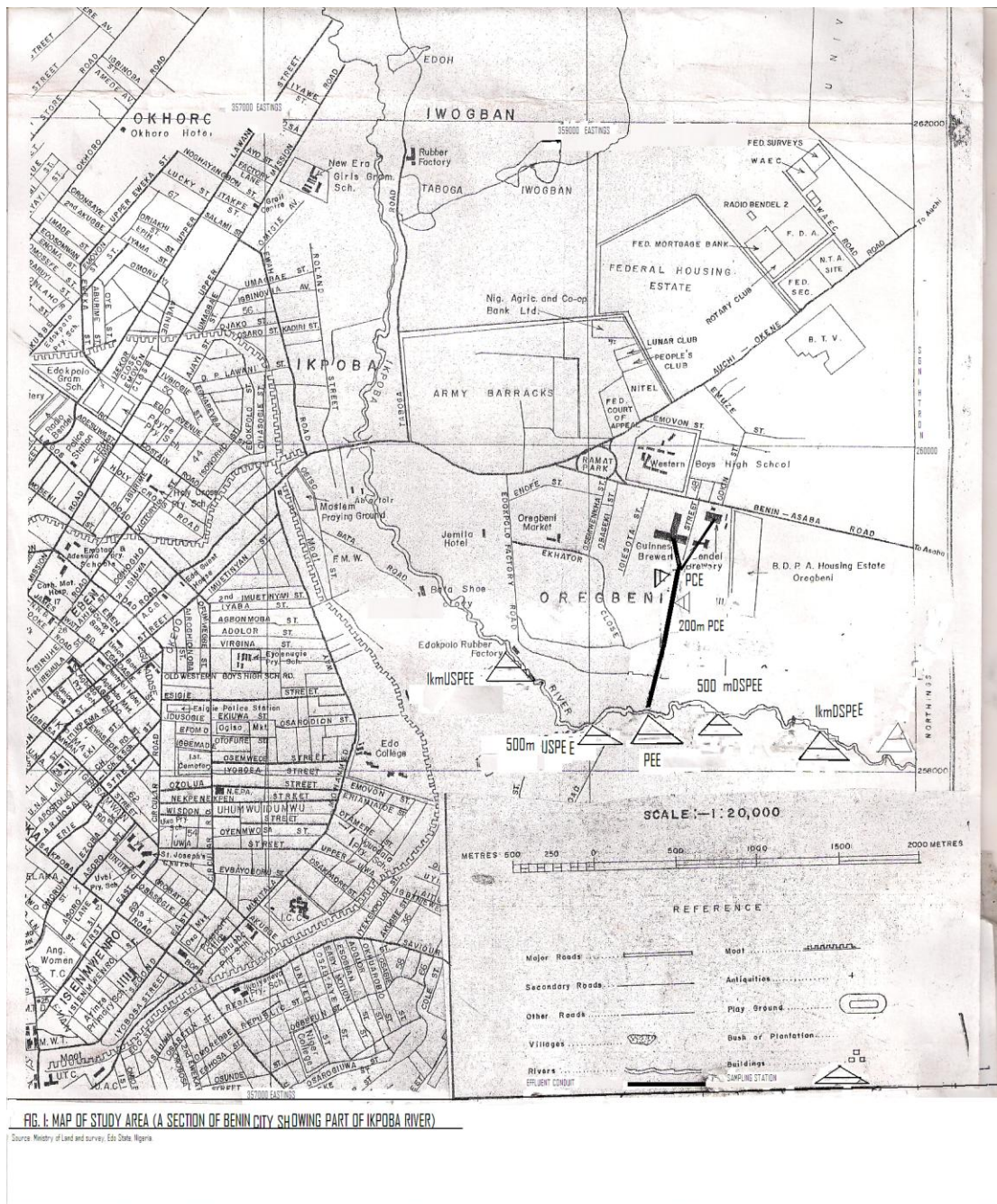
Parameters	Study Area	Control area (Upstream)
pH	6.23 ± 0.40	6.30 ± 0.21
Pb (µg/L)	202 ± 80	183 ± 80
Cu (µg/L)	37 ± 11	28.3 ± 1.9
Cd (µg/L)	21 ± 23	16 ± 17
Cr (Total) (µg/L)	67 ± 64	21 ± 37

Table VI: Pearson 2-tailed correlation of four heavy metals of study area water

	Cu	Cd	Cr	Pb
Cu				
Cd	-0.507*			
Cr	-0.501*	0.978*		
Pb	-0.808**	-0.837**	-0.831**	

*Correlation is significant of 0.05 level (2-tailed)

**Correlation is significant of 0.01 level (2-tailed)



BIOAVAILABILITY OF IRON IN SOME LOCALLY AVAILABLE FRUITS AND VEGETABLES

P.O. Agbaire

CHEMISTRY DEPARTMENT, DELTA STATE UNIVERSITY, ABRAKA

E-mail-patagbaire@gmail.com

Abstract

Iron deficiency is one of the most prevalent micronutrient disorder worldwide. The levels of iron and Ascorbic acid in twenty locally available fruits and vegetables were determined using the flame Atomic Absorption spectrophotometric and spectrophotometric methods respectively. The relative availability of iron in the tested samples was assessed from the ratio of iron to Ascorbic acid (Bioavailability index). The results show that about 71.42% of the fruits show vegetables 1.0 while is 83.33% for bioavailability indices lower than 1.0. These results show that iron in the vegetables are more bioavailable than those in the fruits.

KEYWORDS: Bioavailability index, ascorbic acid, iron availability

INTRODUCTION

Iron is an integral part of many proteins and enzymes that maintain good health. In humans, iron is an essential component of proteins involved in oxygen transport (Anonymous 2009). It is also essential for the regulation of cell growth and differentiation. A deficiency of iron impairs the oxygen supply while excess would cause some sort of poisoning (Anonymous 2009). Iron exist basically in two valence states, these are the ferrous Fe^{2+} and the ferric Fe^{3+} . Dietary irons exist as either the heme or non-heme (Mamta *et al.*, 2006) Heme iron is usually found in animal foods that originally contain haemoglobin while non-heme iron is from plants origin. It has been reported that heme iron are better absorbed than non-heme iron (Miret *et al.*, 2003, Kannan, 2009). Bioavailability of iron is greatly influenced by both dietary enhancers and inhibitors. Ascorbic acid is reported to be the most potent enhance of iron absorption (Halberg *et al.*, 1989; Vijayalashimi *et al.*, 2003; Monsen 1982, and Kannan 2009). It has been reported that in the presence of large amount of ascorbic acid the absorption of iron is greatly enhance due to reduction of Fe^{2+} to Fe^{3+} and subsequent formation of iron (II) ascorbate complex (Vijayalakshmi *et al.*, 2003, Monsen 1982; Ononmhenle *et al.*, 2004). Hence the absorption of iron from vegetables and fruits will therefore depend on the ascorbic acid present. The objective of this study is therefore to examine the relative bioavailability of iron in some fruits and vegetables common in Abraka and its environs.

EXPERIMENTATION

A total of twenty samples were collected from the main market in Abraka. The fruit samples include; Pineapple, Grape, Banana, Cocoa, apple, Tangerine, Orange, Cucumber, Lime, Green pepper, Pawpaw, Tomato, Water melon and Red pepper and the vegetables include Waterleaf, Carrot, Green vegetable, Pumpkin, Bitter leaf and Onion.

Ascorbic Acid Determination

Ascorbic acid content expressed in mg/g was measured using the spectrophotometric method (Pajaj and Kaur 1981). 1g of the sample was measured into a test-tube, 4ml oxalic acid –EDTA extracting solution was added. To this 1ml of orthophosphoric acid was added, followed by 1ml 5% sulphuric acid. Furthermore, 2ml ammonium molybdate was added and finally 5ml of distilled water. The solution as then allowed to stand for 15 minutes. After which the absorbance taken at 760nm with a spectrophotometer. The concentrations of ascorbic acid in the samples were then extrapolated from the standard ascorbic acid curve.

Iron Content Determination:

The iron content was determined by atomic absorption spectrophotometric method after digestion with 20ml concentrated nitric acid and 5ml concentrated sulphuric acid. 5g of sample was weighed and put in a beaker. It was then covered with the digesting acid. The beaker was covered with a watch glass and the solution boiled in a fume cupboard until white fumes of concentrated sulphuric acid started to appear after obtaining a clear solution. The solution was allowed to cool to room temperature and was transferred to a 100ml volumetric flask. Deionised water was added to make to mark. The iron content was then measured with the atomic absorption spectrophotometer-model CY UNICAM 2900.

RESULTS AND DISCUSSION

Table 1: Bioavailability indices of some locally available fruits and vegetables

	Samples common names	Samples scientific names Halize names	Ascorbic acid content (mg/g)	Iron content (mg/g)	Bioavability index
Fruits	Pineapple	Ananas comosus	0.4	0.3	0.8
	Grape	Citrus paradise	0.5	0.2	0.4
	Banana	Musa sapientum	0.3	0.3	1.0
	Cocoa	Theobroma cacao	0.3	0.5	1.7
	Apple	Malus domestica	0.3	0.4	1.3
	Tangerine	Citrus reticulate	0.5	0.1	0.2
	Orange	Citrus sinensis	0.4	0.3	0.8
	Cucumber	Cucumis sativus	0.5	0.3	0.6
	Lime	Citrus aurantifolia	0.7	0.4	0.6
	Green pepper	Capsicum grossom	0.6	0.4	0.7
	Pawpaw	Carica papaya	0.5	0.4	0.8
	Tomato	Lycoperscon cyoprsicum	1.0	1.0	1.0
	Water melon	Citrulus lanatus	1.1	0.3	0.3
	Red pepper	Capsicum connoides	0.9	0.5	0.6
Mean value			0.6	0.4	0.8

Vegetables	Water leaf	Talinum traingulare	0.5	0.3	0.6
	Carrot	Daucus catota	0.7	0.4	0.6
	Green vegetable	Osmanthus heterophyllus	1.6	0.4	0.2
	Pumpkin leaf	Telfera occidentalis	1.4	0.4	0.3
	Bitter leaf	Yamonia amygdalina	0.9	0.4	0.4
	Onion	Allium cepa	0.2	0.3	1.5
Mean values			0.9	0.4	0.6

The table above shows the results of ascorbic acid, iron content as well as the bioavailability indices of some common fruits and vegetables. The results obtained from this work are comparable with other workers (Onomhenle *et al.*, 2004). The ascorbic acid content ranged from 0.2mg/g in *Allium cepa* to 1.6mg/g in *Osmanthus heterophyllus*. The mean iron content in both the vegetable and fruit are the same but the mean bioavailability index in the vegetable is lower than that of the fruit because of the higher ascorbic acid content. This then implies higher iron bioavailability from the vegetables than from fruits. The relatively high bioavailability index of iron in *Telfera occidentalis* may account for the widespread practice of intake of water extract of this vegetable by anaemic patients in most parts of Southern Nigeria.

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**EDTA EXTRACTABLE HEAVY METALS IN SOIL AROUND WASTE DUMPSITES IN
WARRI AND ENVIRONS, DELTA STATE, NIGERIA**

P.O. Agbaire

CHEMISTRY DEPARTMENT, DELTA STATE UNIVERSITY, ABRAKA

E-mail-patagbaire@gmail.com

Abstract

The objectives of this work was to investigate the physioco-chemical properties of soil around refuse dumpsites in Warri and its environs as a well as determine the available heavy metal content of these soils. The extracting solution was 0.05M EDTA and metal measured with the atomic absorption spectrophotometer. The result of the physical properties of soils at dumpsite (DS) showed pH 7.27 ± 0.15 , % sand 46.36 ± 0.03 , % clay 53.61 ± 0.02 , % silt 0.024 ± 0.02 . % TOC 4.98 ± 0.69 while result for 10meters away (10MDS) are pH 7.01 ± 0.23 , % sand 46.37 ± 0.03 , % clay 53.62 ± 0.02 , % silt 0.026 ± 0.02 , % TOC 3.92 ± 0.75 . The results for the CEC are 32.00 ± 10.85 Cmol/kg at DS and 44.26 ± 19.14 Cmol/kg at 10mDS. The results of metal analysis are as follows Fe 0.83 ± 0.36 mg/kg, Pb 0.40 ± 0.28 mg/kg, Cr BDL, Cd 0.24 ± 0.12 mg/kg, Zn 0.68 ± 0.48 g/kg, Cu 0.11 ± 0.08 mg/kg, Ni 0.83 ± 0.43 mg/kg, Mn 0.54 ± 0.24 mg/kg, Co 0.03 ± 0.04 mg/kg for soil around refuse dumpsite (DS). The following are the results for soil at 10mDS. Fe 0.82 ± 0.34 mg/kg, Pb 0.40 ± 0.28 mg/kg, Cr BDL, Cd 0.14 ± 0.11 mg/kg, Zn 0.61 ± 0.56 mg/kg, Cu 0.10 ± 0.09 mg/kg, Ni 0.83 ± 0.85 mg/kg, Mn 0.43 ± 0.55 mg/kg, and Co 0.02 ± 0.03 mg/kg. The student's t-test carried out on the values of metals from dumpsites and 10metres away from dumpsite showed that there was no significant difference. Therefore the soils from the dumpsites could not be said to be contaminated by the waste dumps.

EYWORDS: ETDA extractable heavy metals, dumpsites, Nigeria.

INTRODUCTION

Warri, a city in the Niger-Delta area of Nigeria is an industrialized city. It is located between latitude 5° and 6° N and longitude 5° and 6° N and longitude 5° and 6° E. the area is richly endowed with a number of natural resources. The climate is dominated by two seasons; a long wet season (April – October) and a short dry season (November – March). Annual rainfall is usually in excess of 2600mm and temperature ranges between 23 - 32° C.

The proper waste disposal method has been a serious problem in most cities in Nigeria (Ebong, *et al.*, 2008). Leachates from refuse dumpsites constitute a source of heavy metal pollution to both soil and the aquatic environments (Bamgbose, *et al.*, 2000 and Uba, *et al.*, 2008). In most dumpsite, the waste are burnt in the open and ashes abandoned at the sites. The burning of wastes gets rid of the organic materials and the metals oxidized, thereby leaving the ash richer in metal contents. These metals will dissolve in rain water and leached into soil from where they are picked up by growing plants thereby entering the food chain. Studies have shown that municipal refuse may increase heavy metal concentration in soil and groundwater (Carison, 1976; Albores *et al.*, 2000; Okoronkwo *et al.*, 2005; Okoronkwo *et al.*, 2006). This may affect the host soils, crops and human health (Smith *et al.*, 1996; Nyle and Ray 1999). Thus, the environmental impacts of municipal refuse are greatly influenced by their heavy metal contents. While total heavy contents is a critical measure

in assessing risk of a refuse dumpsite, total heavy metal content alone does not provide predictive insights on the bioavailability, mobility and fate of the heavy metal contaminants (Albores *et al.*, 2004; Srikath and Reddy 1991; Chukuma, 1993). It is however the chemical form or species of the heavy metal that is important factor in assessing their impacts on the environment. Past works on the heavy metals impacts of municipal refuse dumpsites in Nigeria were concerned with total heavy metal only (Bamgbose *et al.*, 2000; Okoronkwo *et al.*, 2006; Abulude 2005). The use of chelating agents such as DTPA and EDTA has been reported as being effective for bioavailability Studies (Lindsay, 1978; Lindsay and Cox, 1985; Kelling *et al.*, 1977; Khaikhaliani, *et al.*, 2006; and Knox and Adriano, 2000). The objective of the work therefore was to investigate the bioavailable heavy metals using ethylenediaminetetraacetic acid (EDTA).

MATERIALS AND METHODS

Soil sample were collected in the month of January. Five domestic dumpsites were identified in Warri and its environs. The dumpsites are Okere market dumpsite, Ekpan, NNPC/DDPA, NNPC Housing Complex and Refinery Road Dumpsite. Top soils were collected from each site and two points were taken. One on dumpsite and the other 10metres away. At each point composite samples of soils were collected with the aid of a soil auger. Samples were then stored in well labeled polythene bags and taken to the laboratory for analysis. On getting to the laboratory, soil were air dried for about 2 weeks, crushed and passed through a 2mm sieve and them stored for until required for analysis (Nwajei *et al.*, 2007; Ebang *et al.*, 2008).

Soil pH 1:2w/v soil to water suspension (Reeuwijk 1995) was used for pH measurement. 10g of the air dried soil was weigh into a 50ml beaker and 20ml of distilled water added. The mixture was allowed to stand for 30minutes with occasional stirring with a glass rod. The electrodes of the calibrated pH meter were then inserted into the partly settled suspension and the pH of the soil measured.

Organic Matter Determination: the soil samples were ground and passed through a 0.5 mm sieve after which they weighed in duplicate and transferred to a 250 ml Erlenmeyer flask. 10ml of IM potassium dichromate was pipetted into each flask and swirled gently to disperse the soil, followed by addition of 20 ml concentrated sulphuric acid. The flask was swirled gently until soil and reagents were thoroughly mixed. The mixture was then allowed to stand for 30 minutes on a glass plate. 100 ml distilled water was added followed by addition of 3 – 4 drops of ferroin indicator, after which it was titrated with 0.5 N ferroin sulphate solution. A blank titration was similarly carried out.

The percentage organic carbon is given by equation

$$\frac{M_1 e_1 K_2 Cr_2 O_7 - M_2 e_2 FeSO_4 \times 0.0031 \times 100 \times F}{\text{Mass (g) of air dried soil}}$$

F = Correction factor (1.33)

M₁ = Mole of k₂ Cr₂ O₇

e₁ = Volume of k₂ Cr₂ O₇

M₂ = Mole of FeSO₄

e₂ = Volume of FeO₄

% Organic matter in the soil = % organic carbon x 1.729 (Anderson and Ingram 1993).

Soil Particle Size: This was measured by the Bouyoucos method (1962) using sodium hexametaphosphate as the dispersing agent after the destruction of organic matter with hydrogen peroxide.

Exchangeable Bases: Exchangeable bases were extracted with neutral NH_4OAc (Rhoades 1982) and then measured using atomic absorption spectrophotometer.

Heavy Metal Determination: Extraction for available heavy metal was done using 0.05M sodium salt of EDTA (Khaikhahani et al, 2006; Uba 2008). Metals were then analyzed using Varian Spectra 220 atomic absorption spectrometer at wavelength specific to each metal.

RESULTS AND DISCUSSION

Table 1A: Physico- Chemical Properties of soils at dumpsites

SITES	PH	TOC	PSD		
			% Sand	% Clay	%Silt
Okere	7.16	5.80	46.35	53.60	0.05
Ekpan	7.13	4.20	46.40	53.60	0.00
NNPC-DDPA	7.36	4.50	46.35	53.65	0.00
NNPC-SSC	7.22	4.80	46.35	53.60	0.05
Refinery Road	7.48	5.60	46.33	53.65	0.02
Mean	7.27	4.98	46.36	53.61	0.02
± SD	0.15	0.69	0.03	0.02	0.02

Table 1B: Physioco-chemical Properties of soils 10meters away from dumpsite

SITES	PH	TOC	% SAND	%CLAY	%SILT
Okere	7.06	4.60	46.40	53.60	0.04
Ekpan	7.06	3.20	46.40	53.60	0.00
NNPC-DDPA	7.33	3.10	46.35	53.65	0.04
NNPC-SSC	6.88	4.00	46.35	53.64	0.01
Refinery Road	6.72	4.70	46.36	53.60	0.04
Mean	7.002	3.92	46.37	53.62	0.03
± SD	0.23	0.75	0.03	0.02	0.02

Table 2A: Concentration of Exchangeable Bases and Cation Exchange Capacity (Cmol/kg) of the soil Around Dumpsite

SITES	Ca	Mg	K	Na	CEC(Cmol/kg)
Okere	7.50	9.00	4.80	3.50	24.80
Ekpan	10.00	7.50	6.40	4.50	28.40
NNPC-DDPA	9.50	7.00	6.80	5.50	28.80
NNPC-SSC	16.50	16.00	9.20	9.50	51.20
Refinery Road	9.00	10.50	4.80	3.00	26.80
Means	10.50	10.00	6.40	5.20	32.00
± SD	3.48	3.62	1.81	2.59	10.85

Table 2B Concentration of Exchangeable Bases and Cation Exchange Capacity (Cmol/kg) of soils 10meters Aways

SITES	Ca	Mg	K	Na	CEC(Cmol/kg)
Okere	19.00	13.00	8.80	16.50	57.30
Ekpan	7.50	7.50	6.40	4.00	25.30
NNPC-DDPA	19.00	15.00	12.40	14.50	60.40
NNPC-SSC	19.00	15.00	14.50	8.00	56.80
Refinery Road	6.50	7.50	4.40	3.00	21.40
Mean	14.20	11.60	9.30	9.20	44.26
± SD	6.58	3.83	4.16	6.09	19.14

Table 3A: Concentration of Heavy metal of soils at Dumpsite (mg/kg)

SITES	Fe	Pb	Cr	Cd	Zn	Cu	Ni	Mn	Co
Okere	1.39	0.54	BDL	0.39	1.42	0.22	1.38	0.89	0.05
Ekpan	0.58	0.75	BDL	0.24	0.88	0.14	1.11	0.65	0.10
NNPC-DDPA	0.47	0.37	BDL	0.16	0.27	0.08	0.56	0.28	BDL
NNPC-SSC	0.78	BDL	BDL	0.10	0.54	0.05	0.28	0.43	0.02
Refinery Road	0.94	0.33	BDL	0.31	0.27	0.08	0.83	0.43	BDL
Mean	0.83	0.40	BDL	0.24	0.68	0.11	0.83	0.54	0.03
± SD	0.36	0.28	BDL	0.12	0.48	0.08	0.43	0.24	0.04

TABLE 3B: Concentration of Heavy Metals of soils 10meters Away from Dumpsite.

SITE	Fe	Pb	Cr	Cd	Zn	Cu	Ni	Mn	Co
Okere	1.30	0.71	BDL	0.31	1.28	0.22	2.22	0.65	0.02
Ekpan	0.66	0.62	BDL	0.08	1.15	0.16	1.11	1.30	0.06
NNPC-DDPC	0.42	0.29	BDL	0.16	0.34	0.05	0.28	BDL	BDL
NNPC-SSC	0.72	BDL	BDL	BDL	0.10	0.03	0.28	0.22	BDL
Refinery Road	1.02	0.38	BDL	0.16	0.20	0.03	0.28	BDL	BDL
Mean	0.82	0.40	BDL	0.14	0.61	0.10	0.83	0.43	0.02
± SD	0.34	0.28	BDL	0.11	0.56	0.09	0.85	0.55	0.03

DISCUSSION

The chemical characteristics of soil from the dump waste site are shown in tables 1A, 2A and 3A while those for 10 meters away from dump waste are shown in tables 1B, 2B and 3B. The comparative mean concentration of properties and standard derivations are also shown in these tables.

Then pH of these soils are close to neutral. This agrees with the findings of Oyelola and Babatunde 2008 and Uba et al, 2008. Heavy metal cations are said to be mobile under acid conditions (Alloway 1996): consequently, the mobility of metal Ions may not be favoured under this near neutral soil condition. The decomposition of organic compounds by the action of micro-organisms increases the level of organic matter. This is evident in the higher TOC content at dumpsite. The organic matter acts as a major absorbent for metals through the formation of chelates and renders them immobile (Alloway 1996). The total mean percentage of organic matter is 4.98 DS and 3.92 10mDS. This is high according to the classification given by Enwezor *et al.*, (1988). The percentage organic matter obtained from this study are higher than those obtained by Bamgbose et al (2000).

The Available heavy metals as extracted through this method which could be said to represent the acid soluble phase in the sequential extract are low as compared with values obtained by Uba et al (2008).

The student's t-test carried out in the values of metals from DS and 10mDS share no significant difference since the calculated values were lower than the table value at 95% confidence level for four degree of freedom. This thus showed that the metal concentration cannot be wholly ascribed to the contribution of waste dump.

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A REVIEW OF TOBACCO SMOKING AS A MAJOR SOURCE OF INDOOR AIR POLLUTION AND ITS EFFECTS.

¹Kpomah B. and ²Kpomah E.D.

¹Department of Chemistry, Delta State College of Sports and Science Education, Mosogar.

²Department of Biochemistry, University of Port Harcourt, Rivers State.

Email: tressurekpomah@yahoo.com, Phone: 08062296458

Abstract

One third of the global population of which 57% men and 43% women smokes cigarette which is derived from the tobacco plant (*Nicotiana*). Tobacco smoking either directly (active) or indirectly (passive) is a major source of indoor air pollution. It is injurious to health because it contains over 4000 toxic and carcinogenic compounds like polycyclic aromatic hydrocarbons (PAH), nitrosamine, alkaloids Lead and Polonium etc, that could cause various ill-healths such as cancer, coronary heart disease (CHD) respiratory impairment, low birth weight in pregnant smokers, erectile dysfunction etc. Tobacco smoking is also a major drain pipe in the finances of smokers, although there are documented benefits associated with smoking such as in the management of Parkinson disease, Alzheimer disease, schizophrenia, ulcerative colitis etc. The disadvantages associated with smoking far outweighs the advantage, hence the fight against cigarette smoking cannot be left in the hands of government alone but non-governmental organisation, co-operate bodies and well spirited individuals should be involved so as to eliminate the only licensed killer in our society.

KEY WORDS: *Nicotiana rustica* , *Nicotiana tobaccum* , *Cigarette smoking* and *carcinogens*.

INTRODUCTION

Cigarette smoking has become fashionable among people of all ages, most especially the youth who use it as a means of showing their 'manhood', highlife and emancipation into civilization. The tobacco plant from which cigarette is derived belongs to the genus *Nicotiana* of which there are about sixty species, but only two of these species can be used for smoking and other human consumption and they are *Nicotiana rustica* and *Nicotiana tobaccum* (Stephen et al, 1999). Tobacco plants are grown in large quantities in countries like Brazil, Cameroon, Cuba, Dominican Republic, Honduras, Indonesia, Mexico, United States of America (USA), Nicaragua, and Nigeria etc. The tobacco leaves are usually harvested when still green and made to undergo curing and fermentation, the tobacco leaves are then rolled and or stuffed into a paper wrapped cylinder (generally less than 120mm in length and less than 10mm in diameter), called cigarette, (Stephen et al, 1999).

Tobacco smoking is the act of burning the dried or cured leaves of the tobacco plant and inhaling the smoke for pleasure or ritualistic purpose for self medication, or out of habit and to satisfy addiction. (Wikipedia, 2007).

Air pollution is the presence in the atmosphere of one or more air contaminants (i.e. dust, fumes, gas, odour, smoke or vapour) in sufficient quantities of such characteristics, and of such duration as

to be or threaten to be injurious to humans, plant or animal life or to property, or which reasonably interfere with the comfortable enjoyment of life or property (Horsefall and Spiff, 2001). Tobacco smoking either directly or indirectly through passive smoking (which is defined as the inhalation of the exhaled and ambient smoke also called second hand smoke or environmental tobacco smoke from one person's cigarette by other people) is a major source of indoor air pollution.

PREVALENCE OF TOBACCO SMOKING:

One third of the global population smokes, 57% of these are men and 43% are women and among young teens (aged 13-15) about one fifth smokes (Medical News Today, 2004). In the United States 48million adults smokes and many cigarettes are smoked in private residence causing a regular release of environmental smoke to roughly 31 million non smokers (11% of the US population) (Nazaroff and Singer, 2004). In Nigeria, there are problems in obtaining reliable working statistical data in every facet of life ranging from population, disease, education, and even on tobacco use, the smoking prevalence in the general population is high considering the 1990 survey of 1270 adults, shows that 24% of men and 7% of women smoke on daily basis, a 32% increase from what it was in 1970 (WHO, 1997). In a survey of 1200 female secondary school students in Anambra State (South Eastern Nigeria), the smoking prevalence was 7.7% and was started at the mean age of 12.6 ± 3.8 years and the number of cigarette smoked per day has a median of two sticks (Ibeh and Ely, 2003). In a Similar survey of 173 out of school youths and apprentices in Ibadan (South West Nigeria) shows that 64.7% claim to have ever smoked, a higher majority of males 73.4% as against female 17.8% (Akinwumi et al, 2006). Also in Ibadan another study of 570 senior secondary school students aged between 15 - 17 randomly selected, 84.5% claimed they smoke in secret premises and other places away from home, 15.3% smoke at home in secret places and 15.7% wherever they like (Olawale and Kehinde 2006). The prevalence might actually be higher because many people (male and female) who smoke are often too shy to own up to the habit based on religious or moral grounds.

TOBACCO SMOKING AND ADDICTION

The addictive nature of tobacco use was first recognized and evident in the mid 17th century when African natives would trade land, livestock and slaves for tobacco. Similarly the Russian Czar prohibited smoking and punished offending subjects by slitting their nostril to discourage the act, yet they did not give up the act (Stephen et al 1999). Nicotine, an element of tobacco smoke is a powerful stimulant and is basically responsible for its addictive nature, although the amount of nicotine inhaled with tobacco smoke is quite small because it is heat labile, but it is however still sufficient to cause physical and or psychological dependence (Wikipedia 2007). The amount of nicotine absorbed by the body from smoking depends on many factors like tobacco type, whether the smoke is inhaled with or without a filter.

When a cigarette is smoked, nicotine reaches the brain from the lungs within seven seconds and the speed with which absorption and distribution occurs is one reason smokers tend to reach out for another cigarette immediately they finish one (Stephen et al, 1999).

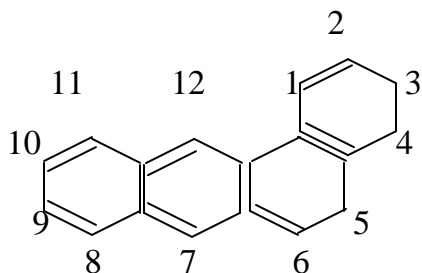
Acute Pharmacological effects of nicotine include: Increase behavioural activity, production of tremor, stimulate vomiting, stimulate the release of antidiuretic hormones, thereby increasing fluid retention, increase in blood pressure and contraction of the heart.

Effect of nicotine overdose may include one or more of any of the followings palpitation, dizziness, Sweating, nausea, vomiting.

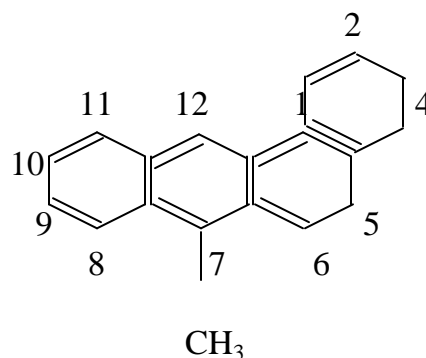
TOXIC, CARCINOGENIC COMPOUND IN CIGARTTE AND HEALTH

Tobacco smoke contains more than 4,000 toxic and carcinogenic compounds (WHO, 2002). The gas phase of cigarette smoke contains a number of volatile nitrosamines including nitrosodimethylamine, nitrosodiethylamine and nitrosopyrrolidine. The particulate matter of cigarette smoke contains polycyclic aromatic hydrocarbon (PAH) which has ten known carcinogens (Glantz and Parmley 1999., Repace 2004). like benzo(a)pyrene and benzo(a)anthracene, as well as nitrosamine derived from tobacco alkaloid nicotine, nornicotine, anabasine and anatribine (Ian and Richard, 1992). The polycyclic aromatic hydrocarbon are of interest as carcinogens (compounds that causes or induced cancer) because their presence in the environment (CBEAP), 1972) is thought to be associated with an increased incidence of cancer of several sites in man and in experimental animals. Members of this class of chemical carcinogens are commonly formed as products of the incomplete combustion of organic materials (Guerin et al, 1987), tobacco been inclusive. The polycyclic aromatic hydrocarbons are usually unreactive, lypophilic and shows only slight solubility in water. Structurally, they are composed of fused benzene rings that may be or not substituted with alkyl groups.

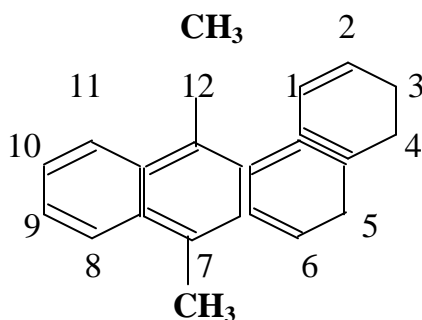
The arrangement of the benzene rings and the sites of substitution have a marked effect on carcinogenic potency (Dippole, 1976) i.e. the addition of methyl groups to the 7 - and 12 - position of the benz(a) anthracene converts a weak carcinogen into a highly active compound.



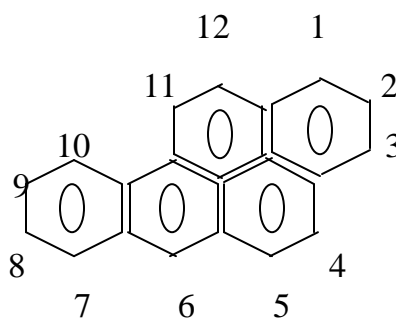
Benz (a) anthracene



7 – methyl benz (a) anthracene



7, 12 – Dimethyl benz (a) anthracene

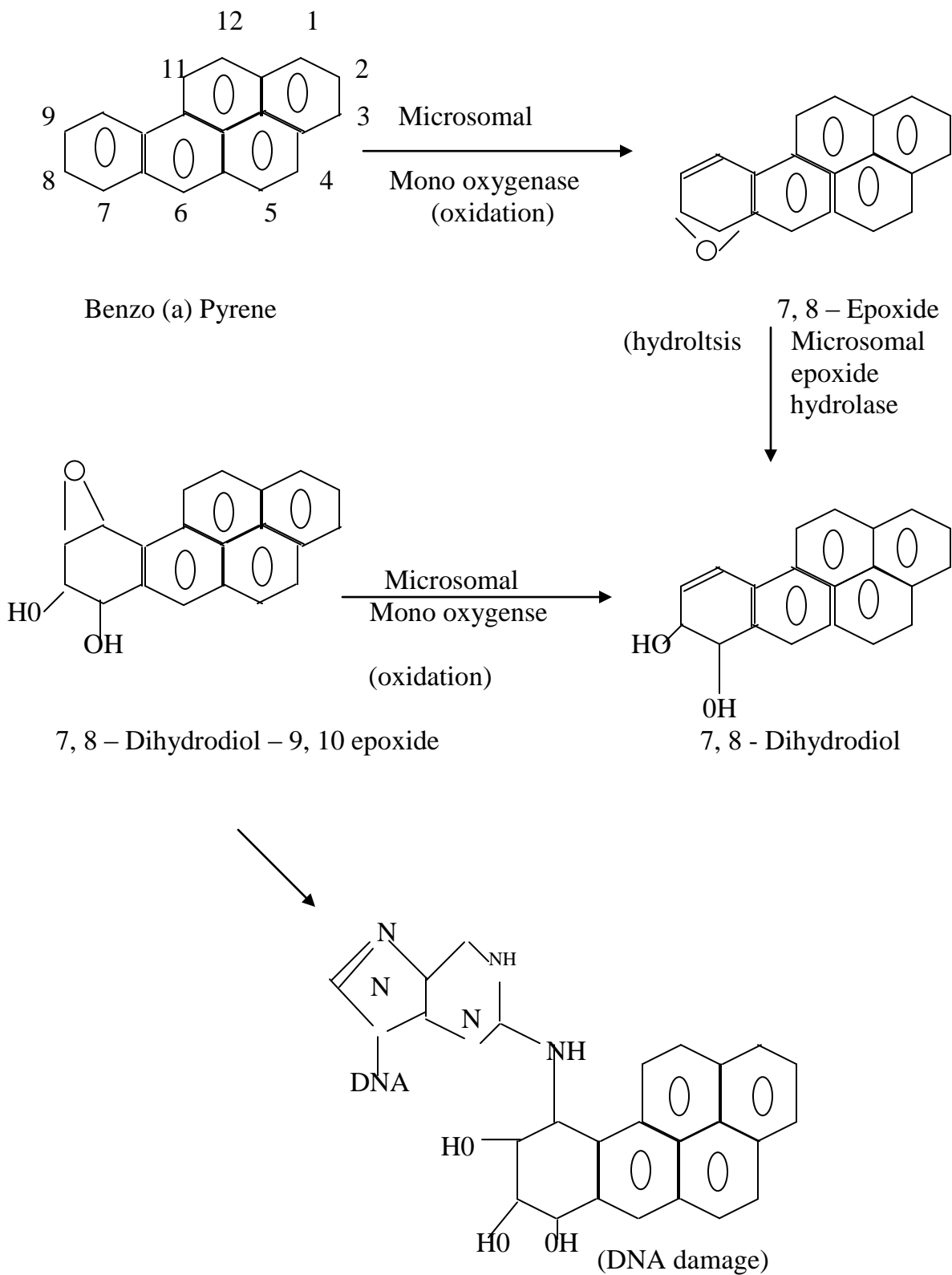


Benzo (a) Pyrene

The first stage in the metabolism of any typical polycyclic hydrocarbon is usually the addition of one

atom of oxygen across an aromatic double bond and this reaction is catalyzed by NADPH - dependent mono oxygenase that are predominantly present in the endoplasmic reticulum of cells (Esterbrook and Lendenkimb, 1979). The simple epoxide so formed can either re-arrange non-enzymatically to the corresponding phenol or be hydrated by the addition of one molecule of water to yield a dihydrodiol and this reaction is catalyzed by epoxide hydrolase (Oesch;1973), the enzymes are also found in the microsomal section of the endoplasmic reticulum, this (electrophile) may react directly with cells (DNA) (nucleophile) or may yet undergo further metabolism to become bound to cells (DNA) covalently, thereby transforming a normal cell into a malignant (Cancerous cell). The most reactive sites in the DNA molecules are the N⁷ and O⁶ of guanine and the N³ and N⁷ of Adenine, however the important site for attack may vary with the particular carcinogen (Timbrel,1991).

MECHANISM OF BIOACTIVATION OF BENZO(A)PYRENE



Tobacco smoke also contains small amount of Lead and Polonium both of which are radioactive carcinogens, Polonium 210 and Lead emits high energy alpha (α) particles that could cause damage to the chromosome (DNA). The radioactive elements in tobacco are accumulated from the minerals in the soil, the radioactivity measured in tobacco varies widely depending on where and how it was grown, this probably explain why tobacco grown in India has lower level of Polonium-210 than that grown in America (Wikipedia, 2007).

Smoking is the single largest preventable cause of disease and premature death and the only licenced killer worldwide, it is a prime factor in heart diseases, stroke and chronic lung disease. It can cause cancer of the lungs, larynx, oesophagus, mouth, bladder, and contributes to cancer of the cervix, pancreases and kidney (WHO, 2002), sudden infant death, low birth weight, continuing growth delay, impaired physical coordination and mental retardation, learning and organizational disorder, erectile dysfunction (impotence), poor vision, poor sense of smell and taste etc.

- **TOBACCO SMOKING AND CORONARY HEART DISEASE (CHD):** Coronary heart disease (CHD) is a term used to identify several cardiac disorders, resulting from inadequate circulation of blood to local area of heart muscles (DHHS, 1988). This deficiency is nearly always a consequence of local narrowing of the coronary arteries by atherosclerosis. The cause of coronary heart disease are multifactor, and tobacco smoking is a key factor because tobacco contain several ingredient that lead to narrowing of blood vessel thereby increasing the likelihood of a blockage. Smoking tends to increase the level of low density lipoprotein (LDL) (Craig et al, 1989), and reduces the level of high density lipoprotein (HDL) (Heiss et al, 1980). Studies have however indicated that low level of high density lipoprotein (HDL) and high level of low density lipoprotein (LDL) increase the channel of developing coronary heart disease (CHD) (Castelli et al, 1977., Millier et al, 1977, Oforofuo and Nwanze, 1994), through the deposition of fatty plaque in the arteries and blood vessel thereby clogging them. The progressive narrowing of the vessel may lead to myocardial infarction (heart attack), hypertension, stroke and sometimes sudden death.
- **SMOKING AND RESPIRATORY AILMENTS:** Heavy smokers have about 25 times more likely chance of dying from lung cancer and chronic obstructive pulmonary disease than non-smokers (Doll et al., 2004). Tobacco smoke contains quite a number of toxic chemical compounds that have a adverse effect on human respiratory system (Laties and Meigan, 1979., Spengler and Sexton, 1983). Studies have also shown that tobacco smoke irritate the upper respiratory tract and the eyes (Hirayawa., 1981) carbon monoxide (CO) is also know to inhibit or cause reduced oxygen intake for normal respiration, similarly carbon monoxide and cyanide (CN⁻) are also know inhibitors of Electron Transport Chain (ETC) at the point of cytochrome oxidase and this can lead to death if not checked immediately.
- **SMOKING AND REPRODUCTIVE HEALTH:** The incidence of impotence is about 85% higher in male smokers when compared to non-smokers (Wikipedia., 2007) and it is a key factor responsible for erectile dysfunction (ED) (Korenman, 2004 and Peate, 2005). Smoking causes impotency by promoting arterial narrowing through clogging by lipids (Low Density Lipoprotein and Triglyceride) (Kendirci et al., 2005). There are increase evidence that the harmful products of tobacco smoke kill's sperm cells (Wikipedia., 2007). Tobacco use is also a factor responsible for spontaneous abortion among pregnant smokers (Ness et al., 1999., Venner., et al, 2004). Tobacco has also been shown to reduce the delivery of oxygen to the foetus (Oncken et al., 2002) through the presence of carbon monoxide, cyanide, and aromatic hydrocarbon. Nicotine and other substances in tobacco smoke also causes reduction in placental blood flow, further causing reduction in oxygen delivery as well as reduction in nutrients to the unborn child thereby causing the risk of low birth weight. Smoking has also been shown to be the largest modifiable risk factor in intrauterine growth retardation

(Wikipedia., 2007).

- **TOBACCO SMOKING AND VISION:** Tobacco smoke has been shown to irritate the eyes (Hirayawa., 1981). The clogging of the blood vessels because of reduced HDL and high LDL many cause reduced blood flow to the eyes (Retina) and this may cause poor vision, development of cataracts and if this is not properly managed may result in blindness.
- **TOBACCO SMOKING AND ORAL HEALTH:** It is a general fact that tobacco causes cancer, coronary heart disease etc. to the general public, but knowledge of tobacco and oral health is a great disbelief for smokers and sometimes the non-smokers alike. Tobacco smoking has a number of well documented side effects which include Tobacco stains and teeth discolouration (Allard et al., 1999., Asmussen and Hassen, 1986), Bad breath (Halitosis). (Christen and Klein, 1997). This bad breath is often directly related to the strength of the tobacco smoked, those with very high concentration of sulphur causes stronger bad breath and the use of breath freshener may not really help because most mint breath fresheners contain high level of sugar and citric acid that may in turn cause dental corrosion (Barylko-Pikielna and Pangbon, 1986), Tobacco smoking may also cause increased calculus deposits, thus potentiating the formation of dental plaques resulting in gum diseases and cavity formation.

Studies have also shown that tobacco is the most significant factor predisposing one to dental implant failure i.e. 4.8% in non-smokers and 11.3% in smokers (Bain and Moy, 1993). Similarly chemical compounds and gases in tobacco greatly affect one's sense of smell, taste and cause series of salivary changes and consequently affecting one's meal, hence one may be tempted to add more salt, pepper or sugar as the case may be and excess of these components in food may be generally toxic and associated with much side effects. Other side effects associated with smoking include

Smokers lip which are cigarette burn on the lips and are generally caused by smoking unfiltered cigarette to the end, which may be regarded as been abnormal, but under the influence of alcohol, this may happen (Mecklenburg et al., 1996), Poor oral wound healing because of the presence of compounds that are vasoconstrictor in tobacco (carbon monoxide) and this greatly reduced blood flow within the month thereby resulting in poor or slow healing of oral injuries as in case of dental surgery.

PERCEIVED HEALTH BENEFIT OF SMOKING

According to the Bible in Ezekiel 47: 12b "The fruit thereof shall be for meat, and the leaf thereof for medicine and according to an English adage "if purpose for something is not known then abuse becomes inevitable" one can therefore say without fear of contradiction that the purpose for Tobacco has been greatly abused because of little or no information regarding it rightful uses.

Tobacco has been found to have some positive health benefits which include the management of the following, Parkinson disease (Fratighoni and Wang, 2002, Allam et al, 2004), Alzheimer disease (Lisa, 2006), Schizophrenia (Compton, 2005., Ripoll et al, 2004), Fibroid (Baron, 1996), Ulcerative colitis (Grun et al, 2000).

SOCIETY AND SMOKING

The society has different perspective on smoking while some see those who smoker as deviants, with psychological problems other see it as a normal way of life but the generality of the public see it as been unacceptable. Christianity, Islam, Judaism and other religious bodies, see cigarette smoking as been unacceptable and sinful against God and hence, they preach against it.

RECOMMENDATION:

Having weighed the pros and cons of cigarette smoking, it is very obvious that the danger associated with its use far exceeds the so called perceived benefits that may be derived from it, for it is not just

injurious to the smokers, it is also a source of financial drain to him or her, at the same time being poisonous to the non-smokers who are exposed to the Environmental Tobacco smoke by virtue of the fact that they find themselves in the same environment as the smokers. Efforts should therefore be intensified by both the Government and Non –Governmental Organization (NGO) to encourage smokers to quit the habit and non – smokers from going into it, considering its addictive nature.

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ULTRACOLD ATOMS IN OPTICAL LATTICES AS QUANTUM SIMULATORS FOR SPIN ORDERING PHENOMENA AND MODELS

Godfrey E. Akpojotor

Department of Physics, Delta State University, Abraka 331001, Nigeria

E-mail: akpogea@delsung.net; Phone: +234 806 660 2677

Abstract

The year, 2010, has been declared the Laserfest to mark the golden celebration of the first demonstration of the light amplification of stimulated emitted radiation (LASER) on May 16, 1960 by Theodore Maiman and his co-workers at Hughes Research Laboratories in Malibu, California, USA. After this first demonstration, the immediate application of the laser was not obvious, consequently it was considered for sometime as a 'solution waiting for a problem.' However, with the development of more compact, reliable and efficient lasers, its application has become proliferated and diverse. Thus part of the Laserfest is to showcase these applications. This paper is dedicated to the Laserfest and it emanates from two presentations made on the Laserfest: the first is on the Laser and its application presented as an invited paper to the plenary session of the 33rd Nigerian Institute of Physics Conference at the University of Ibadan (November 9 – 13, 2010) and the second which is presented at the FOS conference is on the application of laser to the cooling and trapping of ultracold atoms in optical lattices which has become a test laboratory for studying quantum strongly correlated phenomena and models. In particular I will discuss my previous studies of testing the t-U-V-J model which is a spin ordering Hamiltonian using these optical superlattices. Thereafter I will demonstrate the preliminary design of the ultracold atoms in optical kagome lattices and the investigation of the t-U-V-J model in it to provide useful insight into these magnetically frustrated systems.

Introduction

The story of the light amplification of stimulated emitted radiation (LASER) epitomises how theoretical postulation can be developed into a technological harvest. This story began when Albert Einstein postulated the principles of spontaneous and stimulated emission from his fundamental studies on the nature of light in 1917. The basic principle is that simulated emission will occur when a photon interacts with a molecule or atom and causes the emission of a second photon having the same frequency, phase, polarization and direction, thereby creating many nearly identical copies of the incident photon with time. Richard Tolman in 1924 discusses "negative absorption", that is amplification, and explains that the emitted radiation would be coherent with the input radiation. The existence of the phenomenon of stimulated emission and negative absorption was confirmed by Rudolph W. Landenburg in 1928. This first stimulated emission was created in gas giving birth to the creation of microwave amplification by stimulated emitted radiation (MASER) by Charles H. Townes

and Arthur L. Schawlow in 1954. This motivated the search for stimulated emission of photons with shorter wavelengths such as in visible light. The design for such an emission was done independently by Charles H. Townes and Arthur L. Schawlow in 1958 and Gordon Gould in 1959 who is acclaimed to be the first to introduce the term laser in a paper, "The LASER: Light Amplification by Stimulated Emission of Radiation".

The design of the Laser actuated a global race to experimentally demonstrate it. This race was won by Theodore Maiman and his co-workers at Hughes Research Laboratories in Malibu, California, USA. by creating the first working Laser on May 16, 1960. Though Gordon Gould in his design of the laser also included its possible applications such as in spectrometry, interferometry and nuclear fusion, the application of the Laser was not immediately obvious because no one had demonstrated useful applications outside of scientific research as such it was considered for sometime as a 'solution waiting for a problem.'

However, with the development of more compact, reliable and efficient lasers, its application has become proliferated and diverse. And today, it has become a discovery "we cannot do without!" This was made possible because of the laser's distinctive qualities such as its ability to generate an intense, very narrow beam of light of a single wavelength that made its application in science, technology and medicine possible (Townes, 2003). Today, lasers are everywhere: from research laboratories at the cutting edge of quantum physics to medical clinics, supermarket checkouts and the telephone network. The earliest application of the Laser in Medicine was the use of ruby laser to destroy a retinal tumour at Columbia Presbyterian Hospital in December 1961. Other major applications include (for more details: visit Laserfest (2010):

- In 1974, supermarket barcode scanners improved customer checkout times and introduced the public to the first practical application of the laser.
- in 1978, the laserdisc video player launched utilized a He-Ne gas laser first developed right after the ruby laser and was the first true consumer product to include a laser.
- In 1982, the compact audio CD player, the first widely accepted laser-equipped consumer device, was made.

It is pertinent to point out that currently, lasers can create the hottest temperatures on Earth reaching those in the inner core of the sun and also create the coldest temperatures on Earth reaching tens of magnitude colder than liquid nitrogen (which is about 77 K). The latter possibility has helped in the realization of Bose Einstein condensation (BEC) which gave birth to the use of the Laser in cooling and trapping of atoms in optical lattices thereby uniting two formerly distinct aspects of physics: quantum gases from atomic physics (Foot, 2005) and laser theory from quantum optics (Fox, 2006). The optical lattices are artificial crystals of light, that is, a spatially ordered array of potential wells or traps produced by the interference pattern of two or more counterpropagating laser beams. As an insight, a pair of these laser beams in opposite directions (that is, two orthogonal standing waves with orthogonal polarization) will give a one-dimensional (1D) lattice, two pairs in two opposite directions can be used to create a 2D lattice and a similar three pairs in opposite directions will give a 3D lattice. Atoms can be cooled and trapped in these optical lattices so that in a

simple form, the optical lattices look effectively like egg carton where the atoms, like eggs, can be arranged one per well to form crystals of quantum matter (Block, 2008). Thus these quantum crystals can be controlled and manipulated by modifying the frequency, intensity or polarization of the lasers forming them (Lewenstein and Sanpera, 2008). Though the cold atoms in optical lattices was initially used to investigate quantum behaviour such as Bloch oscillations, Wannier-Stark ladders and tunneling phenomena usually associated with crystals in a crystalline solid (Dahan et al., 1996; Wilkinson et al., 1996), it is the theoretical proposal (Jaksch et al., 1998) and consequent experimental realization (Greiner et al., 2002) of the superfluid to Mott insulator (SF-MI) transition which is an important phenomenon in condensed matter physics that has given rise to the possibility of using it as a quantum simulator for phenomena in condensed matter physics. The remaining part of this paper will be focused on this possibility of using the cold atoms in optical lattices as a quantum simulator and it is planned as follows. In the next section, I will review the basic physical principles governing the behavior of atoms in optical lattices. Thereafter I will consider how to simulate quantum models and phenomena using the cold atoms in optical lattices. This will be followed by a brief summary and conclusion.

Trapping atoms in optical lattices

An optical lattice is able to trap atoms because the electric fields of the lasers will induced electric dipole moment in the atom which then interacts with the same electric field of the lasers to produce an effective potential that traps the atom in the optical lattice. The Electric field, E of the standing light wave can be expressed as a product of static spatial and oscillating time-dependent part (Grimm et al., 1999)

$$E(r, t) = \hat{e}E(r)\exp(i\omega t) \quad (2.1)$$

and the induced dipole moment, p oscillating at a driving frequency ω , is

$$p(r, t) = \hat{e}p(r)\exp(i\omega t), \quad (2.2)$$

where $E(r)$ and $p(r)$ are amplitudes and \hat{e} is a unit polarization vector.

The amplitude of the dipole moment is simply related to the field amplitude by

$$p(r) = \alpha(\omega)E(r) \quad (2.3)$$

where $\alpha(\omega)$ is the complex polarizability which depends on the driving frequency.

The field intensity I is related to the field amplitude as

$$I = \epsilon_0 c |E|^2. \quad (2.4)$$

Taking into account Eq. (2.1) – (2.4), the interaction potential of the induced dipole moment and the driving electric field can be expressed as

$$V_{dip}(r) = -\frac{1}{2\epsilon_0 c} \text{Re}(\alpha)I(r). \quad (2.5)$$

The interaction between the induced dipole moment and the driving electric field modifies the energy of the atom which is depicted by the electronic transition within the atom. The difference between the frequency of this transition, ω_0 and the frequency of the laser is called detuning Δ :

$$\Delta = \omega - \omega_0. \quad (2.6)$$

If the laser frequency is less than transition frequency (i.e. $\Delta_t < 0$), the atoms are attracted to potential minima, that is region with maximum electric field intensity and this is known as red detuning while if the laser frequency is greater than the transition frequency (i.e. $\Delta_t > 0$), the atoms are attracted to potential maxima, that is region with minimum field intensity and this is known as blue detuning. Therefore the strength of the optical potential confining the atoms can be increased by tuning the laser intensity, though the atoms can be trapped either in the bright or dark regions of the optical lattice by both types of detuning.

The behaviour of atoms in an optical lattice depends whether they are fermions or bosons. The fermions have half-integer spins and consequently obey the Pauli exclusion principle which states that no two identical fermions can be in the same quantum state at the same time. The physical implication is that fermionic systems will have many energetic particles flying around even as the temperatures goes down to zero as only one particle can occupy the lowest energy. In general, fermions are governed by the Fermi-Dirac distribution (FDD). The bosons, however, have zero or integer spins and consequently do not obey the Pauli exclusion principle. The rules governing the behaviour of photon which is the commonest boson were first given by Satyendra Nath Bose in 1924. Excited by this work, Einstein in the same year extended the rules to other bosons and thereby gave birth to the Bose-Einstein distribution (BED). While doing this, Einstein found that not only is it possible for two bosons to share the same quantum state at the same time, but that they actually prefer doing so. He therefore predicted that when the temperature goes down, almost all the particles in a bosonic system would congregate in the ground state even at a finite temperature. It is this physical state that is called Bose-Einstein condensation.

The Einstein's prediction, however, was considered a mathematical artifact for sometime until Fritz London in 1938 while investigating superfluid liquid helium realized that the phase transition could be accounted for in terms of BEC. This analysis however, suffered a major setback because the helium atoms in the liquid interacted quite strongly. This was why scientists had to move ahead in search of BEC in less complicated systems that would be close to the free boson gas model. Fortunately, the breakthrough came in 1995 when the first BEC was observed in rubidium atoms and this was followed by similar observations in some other cold alkali atoms such as those of lithium and sodium (Anderson et al., 1995; Cornell and Wieman, 2002; Hall, 2003 and Akpojotor and Ojobor, 2008). The achievement of the BEC which won the 2001 Nobel Prize in Physics relied heavily on the then newly developed ability to trap and cool atoms with lasers which was recognized by the Nobel Foundation for the 1997 Nobel Prize in Physics (Metcalf and van der Straten, 1999).

The general belief currently is that almost any kind of atom can be trapped in an optical lattice, but alkali atoms are mostly used due to single valence electron which simplifies description of their behavior in optical lattices. Further, the classification of an atomic isotope to be bosonic or fermionic depends on the number of its constituents: protons, neutrons and electrons. If its number is even, total spin of atom is integer, and the atom is boson while if it is odd, total spin is half integer and the atom is fermion. As bosons the following isotopes are most commonly used: $^{87}_{37}\text{Rb}$, $^{23}_{11}\text{Na}$, $^{39}_{19}\text{K}$, $^{133}_{55}\text{Cs}$; and as fermions: $^{40}_{19}\text{K}$, ^6_3Li , $^{87}_{38}\text{Sr}$.

Simulations with cold atoms in double wells superlattices

To simulate a physical phenomena or model involves mapping it into an alternative physical systems that may be simpler and can easily be manipulated and controlled yet it is described by the same mathematics. The double well (DW) is the simplest experimental set up of optical lattices to simulate physical phenomena and models because the system can be completely controlled and measured in an arbitrary two-spin basis by dynamically changing the lattice parameters (Rey et al., 2007). On the theoretical side, the DW can be considered as two localized spatial modes separated by a barrier and consequently be investigated as a two-mode approximation (Jaksch et al., 1998; Akpojotor and Li, 2008). It is therefore natural that I start the simulation with cold atoms in a DW.

The DW is a 1D optical lattice in which the transverse directions are in strong confinement and thus the motions of an atom in these directions are frozen out (Akpojotor and Li, 2008). To create the double well superlattice which is simply superimposing one lattice on another, we start with a standing wave of period d and depth V_1 (long lattice) and then superimpose on it a counter propagating standing wave with period $d/2$ and depth V_2 (short lattice) as shown in Fig. 1a. The resulting superlattice is a 1D symmetric DW (see Figs 1b and 1c) which can be tilted to obtain an asymmetric DW as shown in Fig. 1d.

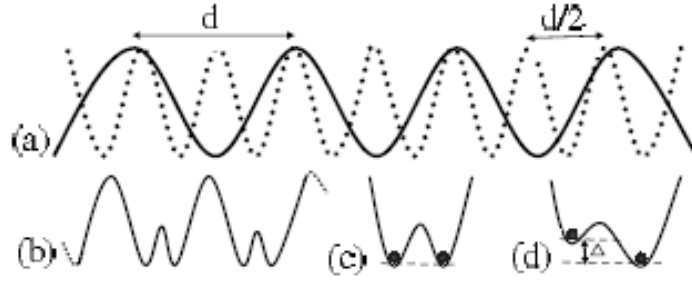


Fig. 1 (a) Two standing waves in opposite directions and with periods d and $d/2$ resulting in (b) a chain of double wells from which we can study (c) a symmetric double well or (d) an asymmetric double well.

The potential seen by the atom in the superlattice DW is (Akpojotor and Li, 2008)

$$V(x) = V_1(x) + V_2 \cos^2(2\pi x/d) \quad (3.1)$$

Therefore, this potential can be manipulated and controlled by varying the depths of the short and long lattices. For example, by increasing the lattice depth of long-lattice V_1 , we could reach from superfluid to Mott-insulator regime, which is convenient for studying the few particles phenomena in a local double-well. And the barrier height of the double-well is controlled by the lattice depth of short-lattice, V_2 . The effective double-well is reached if $V_1 > 4V_2$.

For an atom of mass m trapped in any of the wells corresponding to a filling factor of $1/2$, it will undergo a Josephson oscillation with a frequency of

$$\omega = \frac{\pi}{d} \sqrt{\frac{16V_2^2 - V_1^2}{2mV_2}} \quad (3.2)$$

which obviously depends on not only the lattice depths V_1 and V_2 , but also on the lattice spacing d . Usually, the small lattice spacing is preferred as it leads to a large frequency though this could also be restricted by changing the ratio $V_1/4V_2$. This preference also lead to the use of the recoil energy of the short lattice as the unit of the depths of the optical lattice:

$$E_r = \frac{\hbar^2}{2m\lambda^2} \quad (3.3)$$

where λ is the wave length of the short lattice.

The description so far has been for a 1D symmetric DW (Fig 1c). As discussed above, it can be tilted to obtain asymmetric DW (Fig 1d). The potential bias or the tilt Δ of the DW is introduced by changing the relative phase of the two potentials (i.e. short and long lattices) and this can be realized by applying a magnetic field gradient. Consequently, tuning this field gradient gives the potential difference between the two potential minima of the DW (Trotzky et al., 2008). We can realize the adiabatic and diabatic operations on the tilt of the DW by controlling the increasing speed of the field gradient (Sebby-Strabley et al., 2006).

The starting Hamiltonian for the DW is the two-site version of the Hubbard model (Jaksch et al., 1998; Trotzky et al., 2008 and Akpojotor and Li, 2008; 2009)

$$H_H = \sum_{\sigma=\uparrow\downarrow} -J(a_{\sigma L}^+ a_{\sigma R} + a_{\sigma R}^+ a_{\sigma L}) - \frac{1}{2} \Delta(n_{\uparrow L} - n_{\downarrow R}) + U(n_{\uparrow L} n_{\downarrow L} + n_{\uparrow R} n_{\downarrow R}) \quad (3.4)$$

where $a_{\sigma L,R}^+$ ($a_{\sigma L,R}$) is the creation operator (annihilation operator) for an atom with spin $a(\bar{a}) = \uparrow(\downarrow), \downarrow(\uparrow)$, $n_{\sigma L,R}$ is the corresponding number operator, J (both J and t are used in the literature though the cold matter community seems to prefer J) describes the tunneling rate between the two wells, Δ is the potential bias for the double-well and U is the two-body interaction when two atoms occupy the same site.

Eq.(3.4) is known as the Bose-Hubbard model which for $\Delta = 0$ has a one to one correspondence with the standard Hubbard model with the former being applicable to systems with bosons as the carriers and the latter to systems with fermions as the carriers. The standard Hubbard model proposed in 1963 is universally considered the simplest minimal description of the strongly correlated systems which exhibits some of the most intriguing phenomena in condensed matter physics (Georges, 2004). These include the magnetic, electrical and optical properties and also phase transitions such as in high temperature superconductivity and spin ordering in magnetism. One basic feature common to all these materials is that they are made of electrons that are neither fully itinerant nor fully localized on their atomic sites. Consequently, neither band theory which is successful for many itinerant systems nor localization theory which is used to study systems with localized carriers has been successful for these materials. Therefore, the kinetic part of the Hubbard model is hoped to account for the itinerancy of the carriers while the Coulombic interaction represents the localization of the carriers. In spite of this simplicity, the properties of the Hubbard model have been well determined only in the 1D ($d = 1$) limit where an exact Bethe ansatz solution was obtained in 1968 (Lieb and Wu, 1968) and in the infinite dimension ($d = \infty$) limit where an exact solution has been obtained by Dynamical Mean Field theory (DMFT) in 1989 (Metzner and Vollhardt, 1989). Thus there is no general consensus for the properties at finite dimensions which is the realm for the real materials. It is pertinent to

remark that the model has been investigated with all available theoretical tools that is hoped to be capable of extracting relevant information from the model, as can be found in its rich and vast literature (see Akpojotor (2008) and references therein for more details). One of these tools is to simulate the model with classical computer but this tool has not been successful because of the general problem of the inability to simulate quantum many body problems computationally due to exponential growth of the required space and time resources with the number of particles which often makes the simulation intractable (J'ordan et al., 2008). Therefore, a new tool to investigate the Hubbard model was created when Greiner et al. (2002) experimentally demonstrated the superfluid-Mott transition using cold atoms in optical lattice which was theoretically proposed for the Bose-Hubbard model by Jaksch et al. (1998).

The superfluid to Mott insulator transition is similar to the metal to Mott insulator transition predicted in the standard Hubbard model. To convince the condensed matter community of the possibility of investigating the Hubbard model using cold atoms in optical lattice, the need to use cold fermions in place of the cold bosons used by Greiner et al. (2002) cannot be overemphasized. Therefore, the recent observation of the Mott insulator with cold fermionic atoms by J'ordan et al. (2008) is quite encouraging. The race to achieve metal to Mott insulator transition with cold fermions is still on.

On the contrary, the use of cold bosons in optical lattices has continue to record success so much so that both the theory papers proposed to mimic various condensed matter systems of interest and the number of experiments in which strongly correlated scenarios have be produced have grow significantly that they are now hard to follow the literature. One general consensus, however, is that for spin ordering phenomena and models, there is need to go beyond the Hubbard model either for bosons or fermions (Amadon and Hirsch, 1997; Lewenstein et al., 2007; Trotzky et al., 2008; Akpojotor, 2008a; Akpojotor and Li, 2008; 2009; Liang et al., 2008). For as already explained, achieving the SF-MI transition in the Bose-Hubbard model is simply by varying the potential dept: decreasing the lattice dept will increase the hopping rate so that J dominates while increasing the lattice potential dept will enhance the on-site interaction so that U dominates (Greiner et al., 2002). Introducing a bias potential in the well helps to manipulate the spin ordering of the atoms with very little or no interaction as demonstrated by Trotzky et al., (2008). It follows then that to get nearer the condensed matter scenario wherein the spin ordering is induced by the interaction of the spins, the manipulation via bias potential needs to be replaced by interaction mechanism. In otherwords, we make $\Delta = 0$ to achieve symmetric wells and then introduce appropriate interaction via appropriate laser manipulation to achieve the ordering. Such an extended version of Eq.(3.4) has been suggested in previous studies (Akpojotor and Li, 2008; 2009) by including both nearest neighbor (NN) direct exchange, V and superexchange, J_{ex} interactions (Trotzky et al., 2008) leading to the J - U - V - J_{ex} model:

:

$$H = -J \left[\sum_{\sigma=\uparrow\downarrow} -J(a_{\sigma L}^+ a_{\sigma R} + a_{\sigma R}^+ a_{\sigma L}) \right] + U(n_{\uparrow L} n_{\downarrow L} + n_{\uparrow R} n_{\downarrow R}) + V(n_{\uparrow L} n_{\downarrow R} + n_{\downarrow L} n_{\uparrow R}) - \sum_{\sigma, \bar{\sigma}} J_{ex} (a_{\sigma L}^+ a_{\bar{\sigma} R}^+ a_{\sigma R} a_{\bar{\sigma} L}). \quad (3.5)$$

The DW can be prepared initially either as spin singlet states or spin triplet states so that the common basis states (with a basis state $|L, R\rangle$ denoting the L = left and R = right wells) allowed by the Pauli exclusion principle are $|\uparrow_L \downarrow_L, 0\rangle, |0, \uparrow_R \downarrow_R\rangle, |\uparrow_L, \downarrow_R\rangle, |\downarrow_L, \uparrow_R\rangle, |\uparrow_L, \uparrow_R\rangle, |\downarrow_L, \downarrow_R\rangle$, where the first two are on-site states, the next two are inter-site states and the last two are triplet states (Rey et al., 2007).

Using our highly simplified correlated variational approach (HSCVA) (Akpojotor and Idiodi, 2004 and Akpojotor, 2008a), the exact matrix form of Eq. (3.5) is solved for the DW with fermions to obtain the ground state energy for the singlet and triplet states respectively

$$E_s = -2 \left[\sqrt{\left(\frac{U}{4J} - \frac{V}{4J} - \frac{J_{ex}}{4J} \right)^2} + 1 - \left(\frac{U}{4J} + \frac{V}{4J} + \frac{J_{ex}}{4J} \right) \right] \quad (3.6)$$

$$E_t = 4 \left(\frac{V}{4J} - \frac{J_{ex}}{4J} \right) \quad (3.7)$$

where $U/4J$ is the on-site interaction strength which determines the response of the kinetic energy of the electrons to the varying on-site Coulombic interaction U , $V/4J$ is the NN inter-site interaction strength which determines the response to the varying NN Coulombic interaction and $J_{ex}/4J$ is the NN superexchange interaction strength which determines the response to the varying superexchange interaction J_{ex} . All these quantities are physically dimensionless as they are ratios of the same unit. As expected, the ground state energy for the triplet state is double fold degenerate and this emanate from the up-spins and the down-spins. It has also been shown in our aforementioned previous studies that the transition point, T_p of the ground state energy from E_s (i.e. antiferromagnetic ordering) to E_t (i.e. ferromagnetic ordering) is when

$$\frac{J_{ex}}{4J} > \frac{1}{2} \left[\sqrt{\left(\frac{U}{4J} - \frac{V}{4J} \right)^2} + \frac{1}{2} - \left(\frac{U}{4J} + \frac{V}{4J} \right) \right]. \quad (3.8)$$

The T_p could be sharp, meaning that there is complete cross over from the antiferromagnetic phase to the ferromagnetic phase so that E_t is never equal to E_s for all values of $J/4t$ as depicted in Table 1 for the parameter space: $U/4J = V/4J = 3$; $U/4J = 3, V/4J = -3$; $U/4J = V/4J = 0$; $U/4J = -3, V/4J = 0$ and $U/4J = V/4J = -3$. It has been shown (Akpojotor and Li, 2009) how this sharp T_p can be used to account for the first observed superexchange interaction with cold atoms in optical lattices (Trotzky et al., 2008). Further, prediction of the theoretical values of tunneling parameter and the interaction parameters from extracted data from the experiments of the possible dynamic evolution frequencies,

$$\hbar\omega_{1,2} = \frac{\sqrt{16J^2 + U^2} \pm U}{2}, \quad (3.9)$$

$$\hbar\omega_{3,4} = 2J \left[\sqrt{\left(\frac{U}{4J} - \frac{V}{4J} - \frac{J_{ex}}{4J} \right)^2} + 1 \pm \left(\frac{U}{4J} + \frac{V}{4J} + \frac{J_{ex}}{4J} \right) \right] \quad (3.10)$$

$$\text{and } \hbar\omega_5 = V - J_{ex} \quad (3.11)$$

have been obtained as $J = \frac{1}{2} \hbar \sqrt{\omega_1 \omega_2}$, $U = \hbar(\omega_1 - \omega_2)$, $V = \frac{1}{2}(\omega_3 - \omega_5) - (\omega_1 - \omega_2) + \omega_5$ and $J_{ex} = \frac{1}{2}(\omega_3 - \omega_5) - (\omega_1 - \omega_2) - \omega_5$ (Akpojotor and Li, 2008).

The other possible case for Eq. (3.8) is for the T_P not to be sharp, meaning there is still antiferromagnetic ordering at the onset of that ferromagnetism (i.e $E_t = E_s$) at certain values of $J/4t$ before completely crossing over to the ferromagnetic phase as depicted in Table 1 for the parameter space: $U/4J = 3$, $V/4J = 0$; $U/4J = 0$, $V/4J = 3$; $U/4J = 0$, $V/4J = -3$ and $U/4J = -3$, $V/4J = 0$. This scenario of co-existence is called the mixed state in which the spins are partially polarized and it can be used to investigate two interesting phenomena in condensed matter physics, superconductivity and magnetically frustrated systems.

On-site interaction strength $U/4J$	Nearest neighbour (NN) interaction strength $V/4J$	Nearest neighbour (NN) exchange interaction strength $J_{ex}/4J$	Lowest energy for singlet states E_s at T_P	Lowest energy for triplet states E_t at T_P
3	3	0.3535	10.5857	10.5860
	0	0.0411	-0.1644	-0.1644
	-3	0.0207	-12.0833	-12.0828
0	3	3.0411	-0.1644	-0.1644
	0	0.3535	-1.4143	-1.4140
	-3	0.0411	-12.1644	-12.1644
-3	3	6.0207	-12.0833	-12.0828
	0	3.0411	-12.1644	-12.1644
	-3	0.3535	-13.4143	-13.4140

Table 1: The lowest energies at the transition point, T_P of the antiferromagnetic ordering, E_s and ferromagnetic ordering, E_t for the DW as the on-site interaction strength $U/4t$ and the NN interaction strength $V/4t$ are varied.

Simulations with cold atoms in superlattices beyond the double wells

The defining electromagnetic property of superconductivity is the complete disappearance of the dc electrical resistivity at and below a certain temperature called the critical or transition temperature, T_c . Therefore, the applications of superconductors at room temperature will be diverse and this include its possible use as conductor in the electricity grid which will have far reaching implication for the energy problem. Currently, the highest T_c at normal condition is 134k in the HgBaCaCuO family of the superconducting cuprates. However, there is no consensus theory yet to account for the superconductivity in these materials after more than

two decades of intensive studies of these materials. Recently, I postulated a possible theory to account for the superconductivity of the cuprates (Akpojotor, 2008b) which can be investigated farther using the cold atoms in optical lattices.

The starting point of such an investigation is the observation by Lewenstein et al. (2007) that the spin partially polarized states of cold atoms in optical lattices describes the resonating valence bond (RVB) states that are superposition of states in which random pairs of neighbouring pairs of atoms attains zero total spin. This is because it is this RVB states that was adopted by Anderson (1987) in his proposal that the 2D CuO_2 plane which is generally believed to be the key feature to understanding the high T_c superconducting cuprates, can be reduced to a single band pairing problem. This was confirmed by Zhang and Rice (1988) who then showed that the ground state will be a singlet pairing of the Cu and O and that if liberated as a Cooper pair will lead to superconductivity. I have been able to show the formation of the singlet pairing as the Cooper pair and its propagation within a superexchange interaction (Akpojotor, 2008b). Therefore the design and implementation of the formation and propagation of the Cooper pair using cold atoms in optical lattices can be achieved by improving and extending the study of superexchange interaction in DWs (Trotzky et al., 2008) to other optical structures. This possibility has been boosted by the recent Eckardt and Lewenstein (2010) robust implementation of a quantum simulator for the homogeneous $J\text{-}J_{\text{ex}}$ model with well controlled hole doping, using a sample of ultracold bosonic and fermionic atoms in an optical lattice. It is hoped that the successful demonstration of my theory using the cold atoms in optical lattices will have far reaching implications for understanding high T_c superconductors. One challenge here is how to cool the Fermi gas to temperature below the J_{ex} because if the temperature is not cold enough, thermal fluctuations would destroy the fragile magnetic order present in the ground state (Bloch, 2008).

The magnetically frustrated systems such as the kagome lattice can be studied as the mixed state of the $t\text{-}U\text{-}V\text{-}J$ model because numerical results of the spin $1/2$ system of the Kagome lattice suggest that the energy gap between the ground state and the lowest triplet state, if any, is very small (of the order of $J_{\text{ex}}/20$) and that this gap is filled with low-lying singlets (Waldtmann et al., 1998). As pointed by Lewenstein et al. (2007), these results suggest that the frustrated systems can be described as the RVB states.

The kagome lattice is a 2D frustrated system composed of corner-shared triangles. Since the triangular geometry is believed to have frustrated magnetic ordering (see Fig. 2), the kagome lattices are believed to be magnetically frustrated materials. Frustration here means all the constraints imposed by the Hamiltonian cannot be simultaneously fulfilled (Akpojotor and Akpojotor, 2009).

To design the Kagome lattice with cold atoms in optical lattice, it can be mapped into a 4×4 square lattice as shown in Fig. 2b. It is then easy to see that for Fermi gas there will be a total of 256 singlet states and 240 triplet states while for bosonic atoms there be a total of 136 states. Using the HSCVA, the $t\text{-}U\text{-}V\text{-}J$ Hamiltonian in Eq. (3.5) for Fermi gas will yield an 11×11 matrix which is then solved to obtain the ground state energy at the T_p as the $J_{\text{ex}}/4J$ is increased from zero at $U/4J = 3$ and $V/4J = 0$. The results which is depicted in Fig. 3 clearly show a mixed state that emanates from the spin ordering frustration in the Kagome lattice.

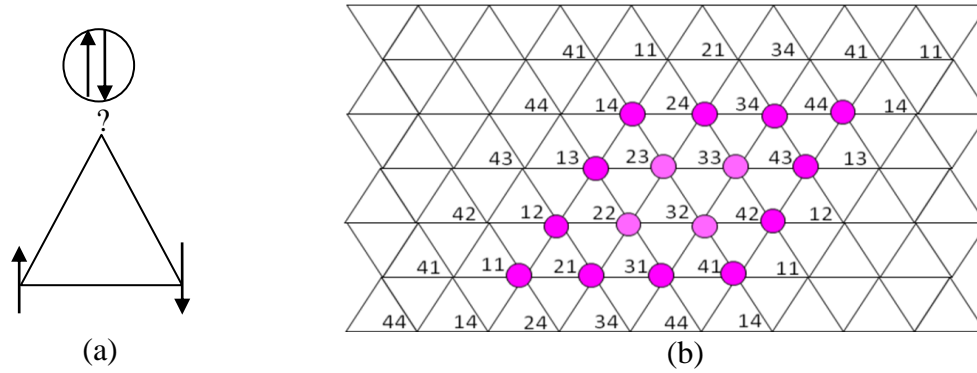


Fig. 2 (Colour online): (a) The triangular lattice as a geometrically frustrated spin system (b) The Kagome lattice formed by the triangular lattices with the round purple circles indicating the possible trapping of cold atoms in a 4 x 4 cluster.

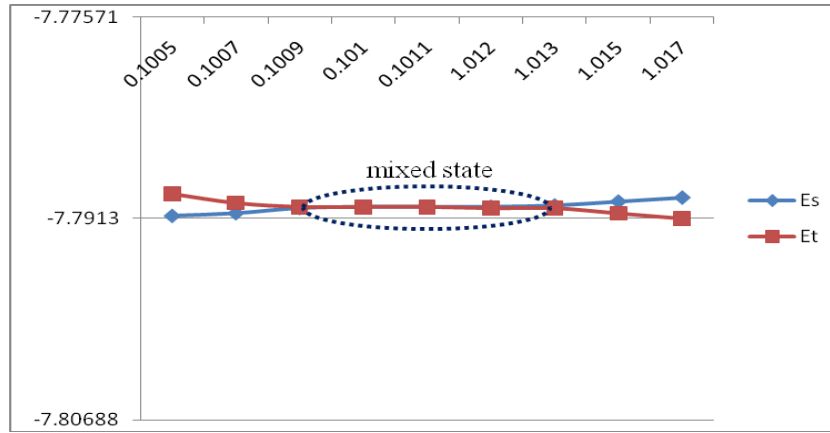


Fig. 3 (Colour online): The change of the ground state energy from antiferromagnetic ordering to ferromagnetic ordering at the transition point, T_p as the superexchange interaction, $J_{ex}/4J$ is increased from zero at $U/4J = 3$ and $V/4J = 0$ for a Kagome lattice of 4 x 4 cluster.

Summary and Conclusion

Summarizing, I have tried to briefly showcase the applications of the laser as part of the golden celebration of its discovery. These applications are so varied and diverse that it has become one discovery that we cannot do without. Therefore, on a speculative postulation, it is hoped that in future there may be hardly any field without the laser applications either directly or indirectly.

I have tried also to give an insight into the application of the lasers for cooling and trapping of atomic gas in optical lattices and therefore refer those interested in a state-of-the-art survey on the active research field of cold atoms in optical lattices to the works of Lewenstein et al. (2007) and Bloch et al. (2008) which also discussed the on-going researches on the application of atomic gases in optical lattices for quantum information processes (Dounas-Frazer et al., 2007). Specifically, the goal of my survey here has been to consider the

possibility of using the cold atom in optical lattices to investigate mixed state of the superexchange J - U - V - J_{ex} model which describes the RVB states that is important in magnetically frustrated systems and also in understanding the superconducting cuprates (Moessner and Sondhi, 2001). Though there are still a number challenges such as cooling to temperatures low enough to prevent thermal fluctuations (Schiro and Fabrizio, 2011) which can destroy the fragile magnetic ordering and also developing techniques to induce the superexchange interactions without a bias potential, the success achieved so far is encouraging. The speculative outlook here is to advance on the techniques to realize more versatile optical lattices such that one can almost control all aspects of the underlying structure and interactions between the atoms trapped in them. This will open the possibility of exploring not only existing phenomena and models in condensed matter physics but also novel ones.

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ON SOME SAMPLING FORMULAS FOR THE TWO - TYPE GALTON-WATSON PROCESSES WITH APPLICATION TO MITOCHONDRIA DNA

J N Igabari¹ and E C Nduka²

¹Dept of Maths and Computer Science, Delta State University, Abraka

²Dept of Maths and Statistics, University of Port Harcourt, Port Harcourt

¹Email: jn_igabari@yahoo.com

Abstract

Many scholars have addressed the so called kin number problem in Galton-Watson populations. One of the most extensive treatments is found in Olofsson and Shaw (2002) with interesting results. The first main result was to provide exact formulas for the mean and variance of proportion of a fixed type in a fixed generation. The second main result was that the sequence of types in the ancestry of randomly sampled particles is a non-homogenous Markov chain where the transition probabilities can be given explicitly, again as a formula. In the present paper, a biological application is considered and it concerns a certain mutation in mitochondria DNA. This application models mutations in Mitochondria DNA as a Galton – Watson branching process of two types (normal and mutant). An exact formula is consequently derived for the mean and variance of the proportion of a particular type in a fixed generation, making use of the probability generating function of the offspring distribution.

KEYWORDS. Galton-Watson process, DNA mutation, Offspring distribution, probability generating function.

INTRODUCTION

Multitype Galton-Watson branching process models have many applications to the study of biological populations. These models are natural tools to study biological systems because they explicitly consider systems of proliferating individuals or particles. The multi-type setting allows individuals to have different probabilistic behaviour depending on their type. In cell and molecular biology, type may for example be genotype, a dichotomous variable indicating mutant vs. wild-type or the number of accumulated mutations.

In a sequence of papers, Waugh (1981), Joffe and Waugh (1982), Shaw (2000), Derrida et al (2000), Oliveira (2002) and Olofsson and Shaw (2002) addressed the so called kin number problem in Galton-Watson populations. They established exact formulas for the probability distributions of family trees of a randomly sampled individual in a fixed generation. One of the most extensive treatment is found in Olofsson and Shaw (2002) with interesting results. The first main result was to provide exact formulas for the mean and variance of proportion of a fixed type in a fixed generation. The second main result was that the sequence of types in the ancestry of randomly sampled particles is a non-homogenous Markov chain where the transition probabilities can be given explicitly as a formula. These results were then applied to what is known as the Polymerase Chain Reaction (PCR), and were used to run simulations of the accumulation of mutations in a fixed number of cycles.

MULTI-TYPE GALTON-WATSON PROCESS

This section contains a brief description of multi-type Galton-Watson process [Mode (1971)].

Suppose that a newborn individual inherits a type from a finite type space $\{0, 1, \dots, r\}$. Given its type, the individual reproduces according to a probability distribution determined by this type. In the application we shall assume that there are two different types and the type space can thus be taken as $\{0, 1\}$. Denote by $X^{(j)}$ the number of offspring of type j and use subscripts to denote types of parents. The offspring distribution is $p_i(k_0, \dots, k_r) = P_i(X^{(0)} = k_0, \dots, X^{(r)} = k_r)$, the probability that an i -type parent gets k_0 children of type 0, ..., k_r , children of type r . The joint probability generating function of $(X^{(0)}, \dots, X^{(r)})$ is defined as

$$\varphi_i(s_0, \dots, s_r) = E_i[s_0^{X^{(0)}}, \dots, s_r^{X^{(r)}}] = \sum_{k_0, \dots, k_r} s_0^{k_0}, \dots, s_r^{k_r} p_i(k_0, \dots, k_r).$$

$$\text{Let } \varphi_i^{(n)}(s_0, \dots, s_r) = E_i[s_0^{Z_n^{(0)}}, \dots, s_r^{Z_n^{(r)}}],$$

be the probability generating function of n^{th} generation, $Z_n^{(0)}, \dots, Z_n^{(r)}$ starting from an ancestor of type i . Now let $s = (s_0, \dots, s_r)$ and let the functions $\varphi : R^{r+1} \rightarrow R^{r+1}$ and $\varphi^{(n)} : R^{r+1} \rightarrow R^{r+1}$ be defined by $\varphi(s) = (\varphi_0(s), \dots, \varphi_r(s))$,

$$\text{and } \varphi^{(n)}(s) = (\varphi_0^{(n)}(s), \dots, \varphi_r^{(n)}(s))$$

Then there is the fundamental recursive relation

$$\varphi^{(n)}(s) = \varphi\left(\varphi^{(n-1)}(s)\right). \quad . \quad . \quad . \quad . \quad (1)$$

or, coordinate-wise,

$$\varphi_i^{(n)}(s) = \varphi_i\left(\varphi_0^{(n-1)}(s), \dots, \varphi_r^{(n-1)}(s)\right) \quad . \quad . \quad . \quad (2)$$

We will use the notation Ψ_n for the probability generating function of $(Z_n^{(0)}, \dots, Z_n^{(r)})$ when there is an arbitrary number of ancestors $(Z_n^{(0)}, \dots, Z_n^{(r)})$, and the notation $\varphi^{(n)}$ for the case of one single ancestor.

Formula for Mean and Variance

The following result was given in Olofsson and Shaw (2002) for the mean and variance of the proportion of type i individuals in the n^{th} generation, conditional on this generation being non-empty and re-echoed in Neves and Moreira (2006). The notation $|Z_n|$ was used for the total number of individuals in the n^{th} generation i.e. $|Z_n| = \sum_{k=0}^r Z_n^{(k)}$.

RESULT – 1 Let u be a vector with all u entries except for a v in the i^{th} position: $u = (u \dots v, \dots u)$, $0 = (0, 0, \dots, 0)$ and denote by Ψ_n the joint probability generating function of $(Z_n^{(1)}, \dots, Z_n^{(r)})$, then

$$E\left[\frac{Z_n^{(i)}}{|Z_n|} \mid |Z_n| > 0\right] = \frac{1}{1 - \Psi_n(0)} \int_0^1 \frac{\partial}{\partial v} \Psi_n(u) \Big|_{u=v=s} ds$$

And

$$\begin{aligned} \text{Var} \left[\frac{Z_n^{(i)}}{|Z_n|} \middle| |Z_n| > 0 \right] \\ = \frac{1}{1 - \psi_n(0)} \int_0^1 -\log s \left(s \frac{\partial^2}{\partial v^2} \psi_n(u) \bigg|_{u=v=s} + s \frac{\partial}{\partial v} \psi_n(u) \bigg|_{u=v=s} \right) ds \\ - \left(\frac{1}{1 - \psi_n(0)} \int_0^1 \frac{\partial}{\partial v} \psi_n(u) \bigg|_{u=v=s} \right)^2 \end{aligned}$$

For proof of these results, see Olofsson and Shaw (2002).

Application to Mutations in Mitochondria DNA

Focus here shall be on a particular type of mutation in mitochondria DNA, which causes a deletion of about one third of the mitochondria genome leading to the production of DNA molecule that is significantly smaller than normal (mutant).

The population of mitochondria DNA is modeled as a two-type process where the types are 0 (normal) and 1 (mutant). A normal can give birth to either two normals or, if there is a mutation, one normal and one mutant. The later happens with probability λ and we refer to λ as the mutation rate. Mutants can only give birth to mutants. A DNA molecule may also die without reproducing (so called mitochondria turnover). Let the survival probabilities be p and q for normals and mutants respectively. This gives the following offspring distributions:

$$P_0(0,0) = 1 - p, \quad P_0(2,0) = p(1 - \lambda), \quad P_0(1,1) = p\lambda, \quad P_0(0,2) = 0 \quad (3)$$

$$\text{for normals and } P_1(0,0) = 1 - q, \quad P_1(0,2) = q, \quad P_1(2,0) = 0, \quad P_1(1,1) = 0$$

for mutants. This gives the joint probability generating functions

$$\varphi_0(u, v) = 1 - p + p\lambda uv + p(1 - \lambda)u^2 \quad (4)$$

$$\text{and } \varphi_1(u, v) = 1 - q + qv^2 \quad (5)$$

The proportion of mutants in the n th generation is

$$\frac{Z_n^{(1)}}{Z_n^{(0)} + Z_n^{(1)}}$$

and we can use Result - 1 to compute its mean and variance. For now, we assume that the population is started from one normal ancestor, that is $[Z_n^{(0)}, Z_n^{(1)}] = (1, 0)$.

To apply Result - 1, note that in this case $\psi_n = \varphi_0^{(n)}$ and by (4.79) we get

$$\psi_n = \varphi_0^{(n)} = \varphi_0(\varphi_0^{(n-1)}, \varphi_1^{(n-1)}) = 1 - p + p\lambda \varphi_0^{(n-1)} \varphi_1^{(n-1)} + p(1 - \lambda) (\varphi_0^{(n-1)})^2 \quad (6)$$

$$\text{and } \varphi_1^{(n)} = \varphi_1(\varphi_0^{(n-1)}, \varphi_1^{(n-1)}) = 1 - q + q (\varphi_1^{(n-1)})^2 \quad (7)$$

Differentiating with respect to v gives

$$\begin{aligned} \frac{d}{dv} \varphi_0^{(n)}(v) &= p\lambda \left(\frac{d}{dv} \varphi_0^{(n-1)} \varphi_1^{(n-1)} + \varphi_0^{(n-1)} \frac{d}{dv} \varphi_1^{(n-1)} \right) + \\ &2p(1 - \lambda) \varphi_0^{(n-1)} \frac{d}{dv} \varphi_0^{(n-1)} \end{aligned}$$

and

$$\frac{d}{dv} \varphi_1^{(n)} = 2q \varphi_1^{(n-1)} \frac{d}{dv} \varphi_1^{(n-1)}$$

Recall that we are interested in

$$\frac{d}{dv} \varphi_0^{(n)}(u, v) \bigg|_{u=v=s}$$

And with the notations

$$F_n(s) = \varphi_0^{(n)}(s, s), \quad f_n(s) = \frac{d}{dv} \varphi_0^{(n)}(u, v) \Big|_{u=v=s}$$

$$G_n(s) = \varphi_1^{(n)}(s, s), \quad g_n(s) = \frac{d}{dv} \varphi_1^{(n)}(u, v) \Big|_{u=v=s}$$

We get the following recursive scheme:

$$F_n(s) = 1 - p + p\lambda F_{n-1}(s)G_{n-1}(s) + p(1-\lambda)F_{n-1}^2(s) \quad (8)$$

$$f_n(s) = p\lambda(f_{n-1}(s)G_{n-1}(s) + F_{n-1}(s)g_{n-1}(s)) + 2p(1-\lambda)F_{n-1}(s)f_{n-1}(s) \quad (9)$$

$$G_n(s) = 1 - q + qG_{n-1}^2(s) \quad (10)$$

$$g_n(s) = 2qG_{n-1}(s)g_{n-1}(s) \quad (11)$$

With the initial conditions ($n = 1$) we have the following:

$$F_1(s) = \varphi_0^{(1)}(s, s) = 1 - p - ps^2 \quad (12)$$

$$f_1(s) = \frac{d}{dv} \varphi_0^{(1)}(u, v) \Big|_{u=v=s} = p\lambda s \quad (13)$$

$$G_1(s) = 1 - q + qs^2 \quad (14)$$

$$g_1(s) = 2qs \quad (15)$$

Note that $f_1(s)$ is not the derivative of $F_1(s)$ with respect to s , but rather the derivative of $\varphi_0(u, v)$ with respect to v evaluated at the point $(u, v) = (s, s)$. To compute $\varphi_0^{(n)}(0)$ we use the recursion formula for F_n and G_n at $s = 0$ to obtain the following results:

$$\varphi_0^{(n)}(0) = F_n(0) = 1 - p \quad (16)$$

$$\text{And } \varphi_1^{(n)}(0) = 1 - q \quad (17)$$

Now to obtain the proportion of normals and mutants respectively in the n th generation, we now use result 1 to get

$$\begin{aligned} E \left[\frac{Z_n^{(i)}}{|Z_n|} / |Z_n| > 0 \right] &= \frac{1}{1 - \varphi_0^{(n)}(0)} \int_0^1 \frac{\partial}{\partial v} \varphi_0^{(n)}(u, v) \Big|_{u=v=s} ds \\ &= \frac{1}{1 - \varphi_0^{(n)}(0)} \int_0^1 f_n(s) ds \\ &= \frac{p\lambda^n}{1 - (1-p)} \int_0^1 s ds \\ &= \frac{1}{2} \lambda^n \text{ for normals.} \end{aligned} \quad (18)$$

This tends to zero as n becomes very large.

For the variance formula, we get a similar recursion scheme where second derivatives are included. For the initial conditions, note that

$$\frac{d^2}{dv^2} \varphi_0(u, v) = 0 \quad (19)$$

and

$$\frac{d^2}{dv^2} \varphi_1(u, v) = 2q \quad (20)$$

4.7.4 Discussion

We stated results about the mean and variance of the proportion of types in a fixed generation in a multi-type branching process. Also, transition probabilities for the sequence of types from a randomly sampled particle in a fixed generation could be obtained using the Markov property. It is important to

note that the formulas are exact and in finite time (fixed generation). This application was to the accumulation of particular deletion in human mitochondria DNA. The branching process considered has two types, normal and mutant. We have used the method of generating functions to model this process and to show that in the long run the normals become extinct under the given hypotheses. Thus, as time goes on, the proportion of mutants increases steadily to 1 so the only stable limiting distribution has probability 1 for mutant and 0 for normal.

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DERIVATION OF A NEW MONKS IMMERSION OF GRASSMANN MANIFOLDS USING GROEBNER BASES

O. Ighedo and H. I. Ojarikre

Department of Mathematics and Computer Science, Delta State University, Abraka

Abstract

Grassmann manifolds are applicable in wireless communications with multiple antennas at the transmitter and receiver, in making of sheets of paper and in making of automobiles. In this paper, for $i = 4$, we establish that $G_{2,2^i-3}$, immerses in $R^{2^{i+2}-15}$ which is an extension of the work of Monks (2001), that for $i = 3$, $G_{2,2^i-3}$, immerses in $R^{2^{i+1}-15}$. This is derived by using the theory of Postnikov towers with the application of Groebner bases for the ideal $I_{2,2^i-3}, I_{2,2^i-4}$.

Key-Words: Grassmann Manifolds, Immersion, Homotopy lifting property, Cohomology ring and Groebner bases

1.0 Introduction

Grassmann manifolds are very important examples of manifolds in mathematics and Physics. These manifolds were introduced by Gunther Grassmann in 1839 when he studied tides and presented special analysis based on vectors. In areas of applications, it was shown by Pawelczy (1997) that Grassmann manifolds were used in the formulation of Gauge fields in terms of a string theory. Also Fujii (2002) used Grassmann manifolds in the application of quantum computation and its efficiency problems. Due to the importance of Grassmann manifolds, extensive research work is being done on it. For instance, Akhmet'ev et al (2003) studied various examples of immersed codimension 1 manifolds which are studied from the view points of possible combinations of the Euler characteristics of the submanifolds of multiple self intersection points. Korbas (2005) offered a new way of deriving bounds for the cup-length of Poincare spaces over fields. He outlined a general research program based on this program. Helmke et al (2007) studied the problem of pole assignment for symmetric and Hamiltonian transfer functions. They gave a necessary and sufficient condition for pole assignment by complex symmetric output feedback transformation. Ighedo & Atonuje (2009) gave an extension of the formalism of Monks (2001), a $G(k,n)$ method. We constructed the space to guarantee that for $i \geq 3$, the space $G_{2,2^i-5}$ does not immerse in $R^{2^{i+1}-3}$ using Groebner bases.

Definition 1:

A topological manifold of dimension n is a second – countable Hausdorff space, each point of which has a neighbourhood homeomorphic to the Euclidean n -space R^n .

Definition 2:

By Grassmannian manifold, $G(k,n)$, we mean the space of all k -dimensional subspaces of an n -dimensional vector space.

Definition 3:

An immersion is a special non-singular map from one manifold to another such that at every point in the domain of the map, the derivative is an injective linear map.

Definition 4:

A map $p:E \rightarrow B$ is said to have the homotopy lifting property with respect to a space X if given a homotopy

$$\begin{array}{ccc} X & \xrightarrow{f} & E \\ & \searrow F & \\ \downarrow i_0 & G & \downarrow p \\ X \times I & \rightarrow & B \end{array}$$

$G:X \times I \rightarrow B$ and a map $f \sim X \rightarrow E$, there exists a homotopy

$F_0:X \times I \rightarrow E$ with $f = F_0$, and $p \circ F = G$ (F is said to be a lifting of G).

Definition 5:

Let $H^*(X;R)$ denote the external direct sum $\bigoplus H^i(X;R)$. The cup product operation makes this group into a ring with a unity element. It is called a cohomology ring of X with coefficients in R .

Definition 6:

Let $I \subseteq Z_2[x_1, \dots, x_k]$ be a non-zero ideal. Let $LT(I)$ denote the set of monomials which are leading terms of I . Fix a monomial order. A finite subset $G = \{g_1, \dots, g_t\}$ of an ideal I is said to be a Groebner bases for I if $\langle LT(g_1), LT(g_2), \dots, LT(g_t) \rangle = \langle LT(I) \rangle$.

2.0 MONK'S DERIVATION

Let M^m denote an n -dimensional paracompact Hausdorff smooth connected manifold. Define $Imm = \min\{j: M^m \text{ immerses in } R^j\}$.

Theorem 2.1;

With respect to the ordering $<$, and for $i \geq 3$,

$A_1) \{\bar{w}_{2^i-2}, \bar{w}_{2^i-1}\}$ is a reduced Groebner basis for $I_{2,2^i-3}$

$A_2) \{\bar{w}_{2^i-3}, \bar{w}_{2^i-2}, \bar{w}_{2^i-1}\}$ is a reduced Groebner bases for $I_{2,2^i-4}$.

Hence we have the following results:

Corollary 2.1.1:

A₃) A vector Space basis for $H^*(G_{2,2^{i-3}}; Z_2)$ is the set of all monomials $w_2^b w_1^a$ such that $a < 2^i - 1$ and $b < 2^{i+1} - 1$. The product structure is completely determined by the relations

$$w_1^{2^i-1} = 0 \text{ and } w_2^{2^{i-1}-1} = \sum_{j=0}^{i-2} w_2^{2^j-1} w_1^{2^i-2^{j+1}} \quad (2.1)$$

A₄) A vector space basis for $H^*(G_{2,2^{i-4}}; Z_2)$ is the set of all monomials $w_2^b w_1^a$ such that $a < 2^i - 1$ and $b < 2^{i-1} - 2$ union with $\{w_2^{2^{i-1}-2}\}$. The product structure is completely determined by the relations $w_1^{2^i-1} = 0$

$$w_2^{2^{i-1}-1} = \sum_{j=0}^{i-2} w_2^{2^j-1} w_1^{2^i-2^{j+1}},$$

$$w_2^{2^{i-1}-2} w_1 = \sum_{j=1}^{i-2} w_2^{2^j-2} w_1^{2^i-2^{j+1}+1} \quad (2.2)$$

Theorem 2.2 (Hirsch 1959);

The following are equivalent

- C₁)** M^m immerses in R^{m+p}
- C₂)** M^m has a normal bundle V which is a p -plane bundle.
- C₃)** There is a lifting V_p of the classifying map of the stable normal bundle of M^m .

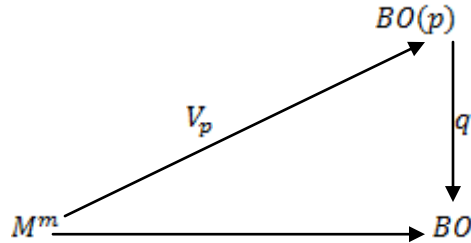


Fig:2.1

(Wu's formula) 2.1.2:

$H^*(BO; Z_2) = Z_2[w_1, w_2, \dots]$ namely

$$S_q^k w_m = \sum_{i=0}^k \binom{m+i-k+1}{i} w_{k-1} w_{m+i} \quad (2.3)$$

Lemma 2.1.3:(Borel 1953)

Let $m \geq 0$. Then in $H^*(BO(2); Z_2) = Z_2[w_1, w_2]$ we have

- (i) $S_q^1 w_1^m = \begin{cases} 0, & \text{if } m \text{ is even} \\ w_1^{m+1}, & \text{if } m \text{ is odd} \end{cases}$
- (ii) $S_q^1 w_2^m = \begin{cases} 0, & \text{if } m \text{ is even} \\ w_2^m w_1, & \text{if } m \text{ is odd} \end{cases}$
- (iii) $S_q^1 w_1^m = \begin{cases} 0, & \text{if } m \equiv 0 \pmod{4} \\ w_1^{m+2}, & \text{if } m \equiv 2, 3 \pmod{4} \end{cases}$

(2.4)

$$(iv) \quad S_q^2 w_2^m = \begin{cases} 0, & \text{if } m \equiv 0 \pmod{4} \\ w_2^{m+1}, & \text{if } m \equiv 1 \pmod{4} \\ w_2^m w_1^2, & \text{if } m \equiv 2 \pmod{4} \\ w_2^{m+1} + w_2^m w_1^2, & \text{if } m \equiv 3 \pmod{4} \end{cases}$$

Lemma 2.1.4:

For $i \geq 3$,

$$S_q^2 S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-5}}) = w_2^{2^{i-1}-2} w_1^{2^{i-2}}$$

Proof:

Applying (2.1.1) and the Cartan formular

$$\begin{aligned} (S_q^1(\alpha \cup \beta) &= \sum_j S_q^i(\alpha) \cup S_q^{i-j}(\beta)) \\ S_q^1(w_2^{2^{i-1}-2} w_1^{2^{i-5}}) &= S_q^1 w_2^{2^{i-1}-2} w_1^{2^{i-5}} + w_2^{2^{i-1}-2} S_q^1 w_1^{2^{i-5}} \\ &= 0 \cdot w_1^{2^{i-5}} + w_2^{2^{i-1}-2} \cdot w_1^{2^{i-4}} \end{aligned} \quad (2.5)$$

$$\begin{aligned} S_q^2 S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-5}}) &= S_q^2 (w_2^{2^{i-1}-2} w_1^{2^{i-4}}) \\ &= S_q^2 w_2^{2^{i-1}-2} \cdot w_1^{2^{i-4}} + S_q^1 w_2^{2^{i-1}-2} S_q^1 w_1^{2^{i-4}} + w_2^{2^{i-1}-2} \cdot S_q^2 w_1^{2^{i-4}} \\ &= w_2^{2^{i-1}-2} w_1^2 \cdot w_1^{2^{i-4}} + 0 \cdot 0 + w_2^{2^{i-1}-2} \cdot 0 = w_2^{2^{i-1}-2} w_1^{2^{i-2}} \end{aligned}$$

Theorem 2.2:

For $i \geq 3$,

$$G_{2,2^{i-3}} \text{ immerses in } R^{2^{i+2}-15}$$

Proof

To simplify notation, we let $m = 2^{i-2} - 2$. It suffices to show that the classifying map for the normal bundle lifts to $BO(8m+7)$.

Let A be the Steenrod algebra.

We shall compute the minimal resolution of the A -module.

$$M = H^j(V_{8m+7}; \mathbb{Z}_2) \text{ for } j \leq 8m+10$$

(the top nonzero grading of $H^*(G_{2,4m+5}; \mathbb{Z}_2)$).

In this range the resolution is

$$M \longleftarrow C_0 \longleftarrow C_1 \longleftarrow \cdots$$

Where M has a generator m_7 in

$H^{8m+7}(V_{8m+7}; \mathbb{Z}_2)$, C_0 is a free- A -module on the generator g_7 in grading $8m+7$ mapping in m_7 and C_1 is a free A -module on a generator h_{10} in grading $8m+10$ which maps into $S_q^2 S_q^1 g_7$.

From this information, we obtained the modified Postnikov tower

$$BO(8m+7)$$



$$\begin{array}{ccc}
 & k_{8m+10} & \\
 & E_1 \longrightarrow & K(Z_2; 8m+10) \\
 & \downarrow & \\
 G_{2,4m+5} & \xrightarrow{v} & BO \xrightarrow{w_{8m+8}} K(Z_2; 8m+8)
 \end{array}$$

Fig:2.1

And it is our goal to lift the map v up to $BO(8m+7)$.

To see that it lifts to E_1 , we notice that the map labeled w_{8m+8} is the classifying map for the cohomology class

$w_{8m+8}(BO; Z_2)$. But we have that

$$w(v) = 1 + w_1 + w_2 + w_1^2 + w_1^3 + w_2 w_1^2 \text{ see (Monks 2001)}$$

thus $V^*(w_{8m+8} \circ v^*)$ is zero.

Thus we have a lifting $l: G_{2,4m+5} \longrightarrow E_1$.

To prove that we can lift this map to $BO(8m+7)$, it suffices to show that $l^*(k_{8m+10})$ is zero. But the lifting is not unique.

We can vary $l^*(k_{8m+10}) \in H^{8m+7}(G_{2,4m+5}; Z_2)$ via the relation that produces k_{8m+10} , namely $(S_q^2 + w_2 + w_1^2) S_q^1 w_{8m+8} = 0$

in $H^*(BO; Z_2)$. It is enough to show that

$$l^*(k_{8m+10}) = (S_q^2 + w_2(v) + w_1(v)^2) S_q^1 x$$

For some $x \in H^{8m+7}(G_{2,4m+5}; Z_2)$. But since $w(v) = 1 + w_1 + w_2 + w_1^2 + w_1^3 + w_2 w_1^2$. But $w_1(v) = w_1$ and $w_2(v) = w_2 + w_1^2$.

Thus we want to show that

$$l^*(k_{8m+10}) = (S_q^2 + w_2) S_q^1 x \text{ for some } x \in H^{8m+7}(G_{2,4m+5}; Z_2).$$

The top nonzero class in $H^*(G_{2,4m+5}; Z_2)$ is in grading $8m+10$.

In Wu's formula, this top class is $w_2^{2^{i-1}-2} w_1^{2^{i-2}}$. So either $l^*(k_{8m+10}) = 0$ or else $l^*(k_{8m+10}) = w_2^{2^{i-1}-2} w_1^{2^{i-2}}$, we see that $w_2^{2^{i-1}-2} w_1^{2^{i-2}} = S_q^2 S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-5}})$.

Also by corollary[2.1.1],

$$\begin{aligned}
 w_2 S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-4}}) &= (w_2^{2^{i-1}-1}) w_1^{2^{i-4}} \\
 &= (\sum_{j=0}^{i-2} w_2^{2^j-1} w_1^{2^{i-2j+1}}) w_1^{2^{i-4}} = (w_1^{2^{i-1}-1}) \sum_{j=0}^{i-2} w_1^{2^{i-2j+1}-3} = 0
 \end{aligned}$$

Hence

$$(S_q^2 + w_2) S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-5}}) = w_2^{2^{i-1}-2} w_1^{2^{i-2}} \text{ as required.} \quad (2.6)$$

Thus we can lift to $BO(8m+7)$ which completes the proof.

3.0 DERIVATION;

For $i=4$

$G_{2,2^i-3}$ immerses in $R^{2^{i+2}-15} \Rightarrow G_{2,13}$ immerses in R^{49}

Proof

We will let $d = 2^{i-2} - 4$.

It is enough to show that the classifying map for the normal bundle lifts to $BO(24d + 23)$.

Let T be the Steenrod algebra.

We shall compute the minimal resolution of the T -module.

Let $D = H^j(V_{24d+23}; Z_2)$ for $j \leq 24d + 26$ (the top nonzero grading of $H^*(G_{2,12d+13}; Z_2)$).

In this range the resolution is

$$D \longleftarrow B_0 \longleftarrow B_1 \circ \circ \circ$$

Fig:3.1

Where D has a generator $m_{23} H^{24d+23}(V_{24d+23}; Z_2)$, B_0 is a free A -module on the generator g_{23} in grading $24d + 23$ mapping into m_{23} and B_1 is a free A -module on a generator h_{26} in grading $24d + 26$ which maps into $S_q^2 S_q^1 g_{23}$.

From this information, we obtain the Postnikov tower $BO(24d + 23)$

$$\begin{array}{ccccc} & & k_{24d+26} & & \\ & & \downarrow & & \\ & & E_1 & \xrightarrow{\quad} & K(Z_2; 24d + 26) \\ & & \downarrow w_{24d+24} & & \\ G_{2,12d+13} & \xrightarrow{\quad v \quad} & BO & \xrightarrow{\quad} & K(Z_2; 24d + 24) \end{array}$$

Fig:3.1

And it is our goal to lift the map v up to $BO(24d + 23)$.

To see that it lifts to E_1 , we notice that the map labelled w_{24d+24} is the classifying map for the cohomology class $w_{24d+24} \in H^{24d+24}(BO; Z_2)$. But we have that

$$w(v) = 1 + w_1 + w_2 + w_1^2 + w_1^3 + w_2 w_1^2 \text{ that } V^*(w_{24d+24}) = 0. \text{ So that, the map } w_{24d+24} \circ V^* \text{ is zero.}$$

Thus we have a lifting $l: G_{2,12d+13} \rightarrow E_1$.

To prove that we can lift this map to $BO(24d + 23)$, it suffices to show that $l^*(k_{24d+26})$ is zero. But the lifting l is not unique. We can vary $l^*(k_{24d+26})$ through $H^{24d+23}(G_{2,12d+13}; Z_2)$ via the relation that produces K_{24d+26} , namely

$$(S_q^2 + w_2 + w_1^2) S_q^1 w_{24d+24} = 0, \text{ in } H^*(BO; Z_2). \text{ It is enough to show that}$$

$$l^*(k_{24d+26}) = (S_q^2 + w_2(v) + w_1(v)^2) S_q^1 x \text{ for some } x \in H^{24d+23}(G_{2,12d+13}; Z_2).$$

The top nonzero class in $H^*(G_{2,12d+13}; Z_2)$ is in grading $24d + 26$.

In Wu's formula, this top class is $w_2^{2^{i-1}-2} w_1^{2^{i-2}}$. So either

$$l^*(K_{24d+26}) = 0 \text{ or else } l^*(K_{24d+26}) = w_2^{2^{i-1}-2} w_1^{2^{i-2}}. \text{ we see that } w_2^{2^{i-1}-2} w_1^{2^{i-2}} = S_q^2 S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-5}}).$$

Also by corollary[2.1.1],

$$w_2 S_q^1 (w_2^{2^{i-1}-2} w_1^{2^{i-5}}) = w_2 (w_2^{2^{i-1}-2} w_1^{2^{i-4}}) = (w_2^{2^{i-1}-1}) w_1^{2^{i-4}} = \left(\sum_{j=0}^{i-2} w_2^{2^j-1} w_1^{2^{i-2j+1}} \right) w_1^{2^{i-4}} = (w_1^{2^{i-1}-1}) \sum_{j=0}^{i-2} w_1^{2^{i-2j+1}-3} = 0$$

Hence

$$(S_q^2 + w_2)S_q^1(w_2^{2^{i-1}-2}w_1^{2^i-5}) = w_2^{2^{i-1}-2}w_1^{2^i-2} \text{ as required}$$

Conclusion

Since we can lift to $BO(24d + 23)$, we conclude that
for $i = 4, G_{2,2^i-3}$ immerses in $R^{2^{i+2}-15}$.

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NOISE STIMULATED OSCILLATION IN SOLUTIONS OF STOCHASTIC DELAY DIFFERENTIAL EQUATIONS WITH POSITIVE COEFFICIENTS AND TWO DEAD TIMES

Augustine Omoghaghare Atonuje

Department of Mathematics and Computer science, Delta State University, Abraka, Nigeria.

Abstract:

The paper considers a scalar linear delay differential equation

$$\left. \begin{aligned} x'(t) &= p(t)x(t-r) + q(t)x(t-\sigma), \quad t \geq 0 \\ x(t) &= \phi(t), \quad t \in [-\Gamma, 0] \end{aligned} \right\} \quad (*)$$

having positive coefficients and two deviating arguments. The contribution of multiplicative noise of Ito type to the oscillatory behaviour of solutions of (*) is studied.

It is proved that the addition of noise to (*) would stimulate oscillation in solutions of the resulting stochastic delay differential equation (SDDE) for negative feedbacks. If the noise is deleted, the deterministic system (*) can admit a non-oscillatory solution which is not possible with the stochastic case. Hence the dead times are no longer the sole cause of oscillation.

1.0 INTRODUCTION:

Both deterministic and stochastic delay differential equations are applicable in mathematics, economics, engineering, epidemiology and population dynamics for modeling; active vibration and noise control systems, conveyor belts, metal rolling systems, remote control systems, urban traffic, electronic transmission lines, heat exchangers, population dynamics (taking into account the gestation times), infectious diseases (accounting for the incubation periods), physiological and pharmaceutical kinetics (such as the body reaction to CO₂ in circulating blood), navigation control of ships and aircrafts, machine chatter, etc (See for instance Tobias [1965], Kuchler and Platen [2007], Baker and Buckwar [2003]).

In recent years a lot of attention has been focused on the investigation of the oscillatory and non-oscillatory properties of deterministic delay differential equations with emphasis on obtaining necessary and sufficient conditions for oscillation and in some cases existence of positive solutions (See for instance, Ahmad [2003], Elabbasy et al [2000], Li [1996], Agwo [1999]) and the references therein. In spite of the fact that many paper articles, research monographs and textbooks have been written on the oscillatory properties of classical DDEs, it appears that only a little has been achieved in respect of the contribution of multiplicative noise of Ito type to the oscillatory behaviour of solutions of first order delay differential equations, thus making it an interesting topic to research further. It is well known that oscillation in first order delay differential equations (both deterministic and stochastic) is caused by the presence of long enough delays. {Gopalsamy [1992], Ladde et al [1987], Appleby and Buckwar [2005]}.

The work on this paper is motivated by the following interesting results: In 2005, Appleby and Buckwar [2005] studied the almost sure oscillatory properties of first order stochastic delay

differential equations with a single variable delay. Later Atonuje and Ayoola [2008] focused a great attention on the problem of the effects of multiplicative noise on the oscillatory behaviour of solutions of SDDEs with multi-constant time lags and those with positive and negative coefficients.

The key question which we intend to answer in the present paper is "If the corresponding deterministic (non-stochastic) DDE has a non-oscillatory solution under certain conditions, can this happen in the presence of a multiplicative noise? In particular, what is the contribution of noise to the oscillatory and non-oscillatory behaviour of solutions of stochastic delay differential equations? Or what is the role played by noise in the creation, existence and destruction of oscillations of solutions of first order delay

differential equations with positive coefficients and two time lags? We propose to explain that under certain assumptions, the existence of oscillatory solutions of first order stochastic delay differential equations is not only caused by time delay influence but it is the consequence of the interplay between internal noise and time delays. We do this by employing the formalism in Appleby and Buckwar [2005] and a technique of solution decomposition originally established in Lisei [2001].

The paper is organized in three sections. We begin with introduction in the first section. The second section contains preliminary results and the last section contains the main result. It is our hope that this work will encourage other researchers to direct more attention to the study of the effects of noise perturbation on the oscillation of solutions of feedback processes.

2.0 Preliminary Results::

Definition 1:

A solution $x(t)$ of a DDE defined on an interval $[T_x, \infty)$ and satisfies $\sup\{|x(t)| : t > T\} > 0$ for every $T \geq T_x$ i.e. $|x(t)| \neq 0$ in any infinite interval $[T_x, \infty)$ is said to be a regular or non-trivial solution.

A solution $x(t)$ of a DDE is called a zero solution if $x(t) \equiv 0$ whenever the initial function $\phi(t) \equiv 0$. It is also called an equilibrium solution. This is also true of the solution of the SDDE $X(t)$.

Definition 2:

A regular (i.e. non-trivial) solution $x(t)$ of a DDE is said to be eventually positive if there exists $t_1 > 0$ such that $x(t) > 0$ for $t \geq t_1$. A regular (i.e. non-trivial) solution $x(t)$ of a DDE is said to be eventually negative if there exists $t_1 > 0$ such that $x(t) < 0$ for $t \geq t_1$.

Definition 3:

A non-trivial (or regular) solution $x(t)$ of a deterministic DDE is said to be oscillatory if and only if it has arbitrarily large zeros for $t > 0$, i.e. if there exists a sequence $\{t_n : x(t_n) = 0\}$ of $x(t)$ such that $\lim_{n \rightarrow \infty} t_n = +\infty$. Otherwise it is said to be non-oscillatory.

The definition of oscillation above was introduced in 2005 by Appleby and Buckwar [2005] into stochastic processes as below:

Definition 4:

A non-trivial continuous function $f : [t_0, \infty) \rightarrow \mathbb{R}$ is oscillatory if the set $W_f = \{t \geq t_0 : f(t) = 0\}$ satisfies $\sup W_f = +\infty$. A function which is not oscillatory is said to be non-oscillatory.

This notion is extended to stochastic processes in the following intuitive manner:

A stochastic process $\{X(t)\}_{t \geq 0}$ defined on a probability triple (Ω, F, P) and with continuous sample paths is said to be almost surely (a.s.) oscillatory if there exists $\Omega^* \subseteq \Omega$ with $P[\Omega^*] = 1$ such that for all $w \in \Omega^*$ the path $X(\cdot, w)$ is oscillatory or it is said to be non-oscillatory.

Problem Formulation:

We consider the oscillatory properties of solutions of the first order stochastic delay differential equation SDDE

$$\left. \begin{aligned} dX(t) &= [p(t)X(t-r) + q(t)X(t-\sigma)]dt + \mu X(t)d(t), \quad t \geq 0 \\ X(t) &= \phi(t), \quad t \in [-\Gamma, 0] \end{aligned} \right\} \quad (2.1)$$

With positive coefficient $p(t) \geq 0, q(t) \geq 0$ which are continuous, $r > 0, \sigma > 0$ are positive constant time lags, the initial function $\phi \in C([-\Gamma, 0], \mathbb{R})$, $\Gamma = \max\{r, \sigma\}$ and

$\{B(t)\}_{t \geq 0}$ is a one-dimensional standard Brownian motion. By solution of equation (2.1), we mean a stochastic process $\{X(t)\}_{t \geq 0}$ defined on a probability space (Ω, F, P) which satisfies equation (2.1) as well as its initial function ϕ . We will always compare the oscillatory behaviour of solutions of the SDDE (2.1) with the oscillatory behaviour of solutions of the corresponding classical DDE

$$\left. \begin{aligned} x'(t) &= p(t)x(t-r) + q(t)x(t-\sigma), \quad t \geq 0 \\ x(t) &= \phi(t), \quad t \in [-\Gamma, 0] \end{aligned} \right\} \quad (2.2)$$

By solution of equation (2.2), we mean a function $x(t) \in ([t_0 - \Gamma, \infty), \mathbb{R})$ for all $t \geq t_0$.

Notable characteristics of the sample path of solution of a scalar SDDE (being a stochastic process) are that there are no points at which the path is differentiable. Also it is of unbounded variation in any finite interval; hence one may think that oscillation of solutions of such equations is a generic (not special) concept. Consequently, to prove the existence of oscillatory and non-oscillatory solutions of the SDDE (2.1), we write the solution $X(t)$ of the SDDE in terms of a random non-autonomous linear delay differential equation which admits a continuously differentiable solution $Z(t)$ of the form

$$Z'(t) = - \sum_{i=1}^n H_i(t) Z(t-r_i) \quad (2.3)$$

Where each $H_i(t)$ is a positive random function defined on some subset $\Omega^* \subseteq \Omega$ by

$$H_i(t)(w) = \begin{cases} -be^{-\lambda r_i} e^{(-\mu(B(t)(w) - B(t-r_i)(w)))}, & \text{for } t > \underline{t} \\ -be^{-\lambda t - \mu B(t)(w)}, & \text{for } t \leq \underline{t} \end{cases} \quad (2.4)$$

Where, $\lambda = \left(a - \frac{\mu^2}{2}\right)$, $\underline{t} = \inf\{t > 0 : t - r_i = 0\}$ such that for all $t > \underline{t}$,

$$t - r_i \geq 0 \text{ and } w \in \Omega$$

We see that the H_i depend upon the increments of a standard Brownian motion $\{B(t)\}_{t \geq 0}$. If the deviations in these increments are sufficiently large, oscillation is generated equation (2.3). Next we shall recall for use on a path-wise basis, that is, for each $w \in \Omega$ some extensive and useful results in the oscillatory theory of deterministic delay differential equations which apply to the sample paths of the random delay differential equation (2.3).

The following concerning oscillatory property has been inspirational to several authors in oscillation of DDEs. It is a special case of the result found in Ladas [1997].

Lemma 1:

Assume that $r, \sigma \in (0, \infty)$ and each $H : [0, \infty) \rightarrow [0, \infty)$ is continuous such that

$$\liminf_{t \rightarrow \infty} \int_{t-\Gamma}^t \sum_{i=1}^n H_i(s) ds > \frac{1}{e} \quad \text{and} \quad \liminf_{t \rightarrow \infty} \int_{t-\frac{\Gamma}{2}}^t \sum_{i=1}^n H_i(s) ds > 0 \quad (2.5)$$

Then every solution of

$$Z'(t) = - \sum_{i=1}^n H_i(t) Z(t-r) \quad (2.6)$$

is oscillatory.

We also have results pertaining to non-oscillatory solutions. The following is a special case of the result found in Gopalsamy [1992] (Proposition 1.3.6). It originally appeared in Yan [1987].

Lemma 2:

Equation (2.6) has a non-oscillatory solution if and only if the integral equation

$$x(t) = \sum_{j=1}^m H_j(t) \exp \left[\int_{t-\Gamma}^t x(s) ds \right] \quad (2.7)$$

Has a solution $x(t)$ on $R_T = [T, \infty) \subseteq \mathfrak{R}_+$

Solution Transformation:

In order to investigate the oscillatory behaviour of the solution of the SDDE (2.1) being a stochastic process, we transform the solution $X(t)$ of the SDDE into a conjugation relation with the solution of the random scalar delay differential equation (2.3). This is done by using a bijective random process $\{\eta(t)\}_{t \in \mathfrak{R}}$ with known properties. To this end, we introduce the process with properties as below:

Lemma 3: (Lisei [2001] Theorem 3.2)

The following assertions hold:

H₁: $\{\eta(t, \cdot)\}_{t \in \mathfrak{R}}$ is a continuous $C^{k+1, \epsilon}$ semi-martingale (with $0 < \epsilon < \delta$) such that for all

$w \in \Omega, \quad \mathfrak{R}^d \ni v \rightarrow \eta(t, v) \in \mathfrak{R}^d$ is a C^{k+1} diffeomorphism of

\mathfrak{R}^d and $\{\Gamma(t, \cdot)\}_{t \in \mathfrak{R}}$ is a continuous $C^{k, \epsilon}$ semi-martingale (with $0 < \epsilon < \delta$).

H₂: For all $t \geq s, v \in \mathfrak{R}^d$ and almost everywhere (a.e) $w \in \Omega$

$$\eta(t, v) = \eta(s, v) + \int_s^t M(du, \eta(u, v)) + \int_s^t \Gamma(u, v) du \quad (2.8)$$

where $\Gamma(t, \cdot)$ is a random process.

H₃: The processes $\{\eta(t, \cdot)\}_{t \in \mathfrak{R}}, \quad \{\Gamma(t, \cdot)\}_{t \in \mathfrak{R}}$ are perfectly stationary, that is,

$$\eta(t, v, w) = \eta(0, v, \theta(t, w)) \quad \text{and} \quad \Gamma(t, v, w) = \Gamma(0, v, \theta(t, w)) \quad (2.9)$$

For all $t \in \mathfrak{R}, v \in \mathfrak{R}^d, w \in \Omega$.

We now define a conjugation relation among $X(t, w)$, the solution of the SDDE (2.1), $Z(t, w)$ the solution of the random DDE (2.6) and bijective random coordinate change

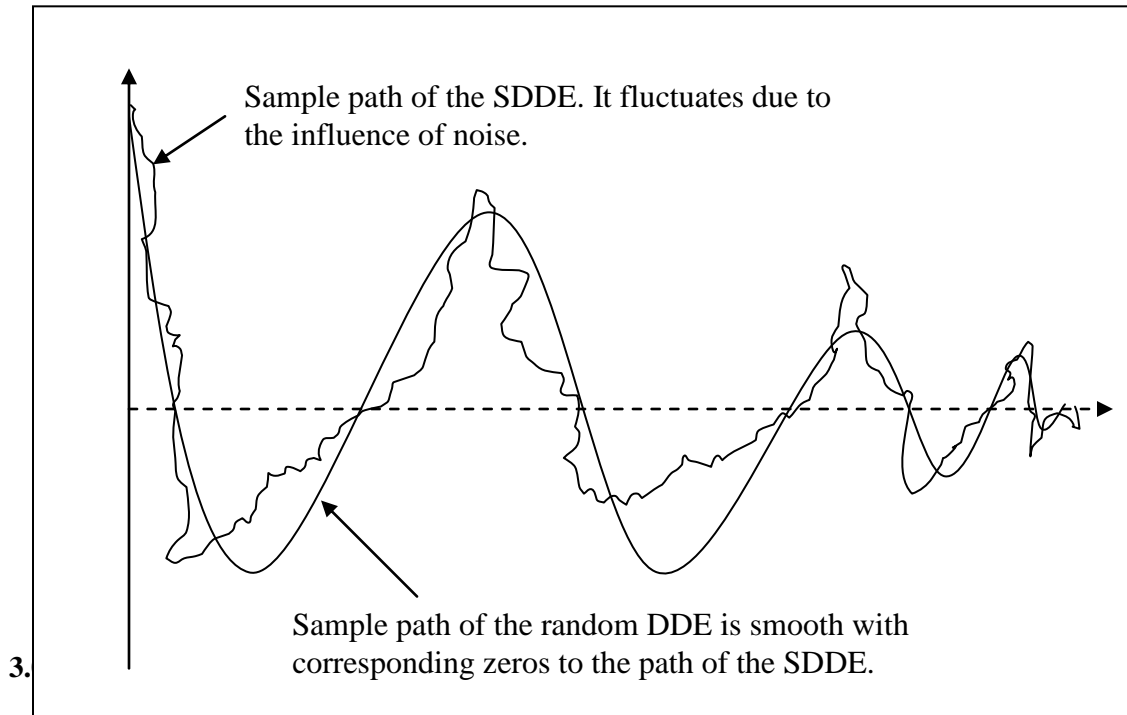
$\{\eta(t, \cdot)\}_{t \in \mathbb{R}}$ with known properties as in Lemma 3 as follows:

$$X(t, \cdot, w) = \eta(0, \cdot, \theta(t, w)) \circ Z(t, \cdot, w) \circ \eta^{-1}(0, \cdot, w) \quad (2.10)$$

For all $t \geq 0, v \in \mathbb{R}^d, w \in \Omega$.

Equation (2.10) builds a conjugation relation between the nowhere differentiable solution X of the SDDE and the continuously differentiable solution Z of the random DDE. The zeros of X correspond to the zeros of Z . By the conjugation relationship, we now analyze the oscillatory properties of the solution of the random DDE (2.6) in order to obtain information about oscillatory behaviour of solutions of the SDDE (2.1). A similar technique of building a product relationship was used by Appleby and Buckwar in 2005. Useful deterministic oscillatory results (as we have in Lemma 1 and Lemma 2 in this case) are now invoked on a path-wise basis to Z so as to study X .

Figure 1: Corresponding zeros of sample paths.



$f(t) = t - \Gamma$ satisfies the hypothesis of Lemma 1, the solution of the SDDE (2.1) is certainly oscillatory in the half interval $[0, \infty)$ for whatever choice of initial function ϕ .

Theorem 1:

Assume that $p < 0, q < 0$ and $0 < r \leq \sigma < \infty$. Then the SDDE (2.1) has an oscillatory solution on $[0, \infty)$ with probability 1 for every choice of continuous initial datum ϕ .

Proof:

By the relationship in equation (2.10) and since $\eta(t)$ is perfectly stationary, the set $W = \{t > 0 : X(t) = 0\}$ can only satisfy $\sup W^* = +\infty$ if and only if the set $W^* = \{t \geq 0 : Z(t) = 0\}$ satisfies $\sup W^* = 0$ following from the definition of oscillation of a continuous functions. Since

$\{X(t)\}_{t \geq 0}$ is defined on the probability triple (Ω, \mathcal{F}, P) , we define for $t \geq 0, w \in \Omega$

$$\sum_{i=1}^N H_i(t, w) = -q(t) \eta(t - r_i, w) \eta^{-1}(t, w).$$

Then each $H_i(\cdot)$ is an almost certainly positive continuous function on $[0, \infty)$. Moreover, Z satisfies the equation

$$\left. \begin{aligned} Z'(t, w) &= -\sum_{i=1}^n H_i(t, w) Z(t - r_i, w), \quad t > 0 \\ Z(t) &= \phi(t), \quad t \in [t_1 - \Gamma, 0] \end{aligned} \right\}. \quad (3.1)$$

Hence it satisfies conditions of Lemma 1 and thus almost certainly oscillatory. Let us suppose that Z is not oscillatory and for the sake of contradiction, equation (2.1) may have an almost sure positive solution $Z(t)$.

Assume that there exists a $t_1 > 0$ such that $Z(t) > 0$ for $t > t_1$ and $Z(t - r) > 0$ for $t > t_1 + r$. Hence $Z'(t) < 0$ for $t > t_1 + r$ and $Z(t) < Z(t - r)$ for $t > t_1 + 2r$.

Introduce $k(t) = \frac{Z(t - r)}{Z(t)}$, for $t > t_1 + 2r$. We see that $k(t) > 1$. Express equation (2.6) as

$$Z'(t) + \sum_{i=1}^n P_i(t) Z(t - r_i) = 0 \quad (3.2)$$

And dividing both sides by $Z(t)$ yields

$$\frac{Z'(t)}{Z(t)} + \sum_{i=1}^n H_i(t) k(t) = 0 \quad \text{for } t > t_1 + 2r \quad (3.3)$$

Integrating equation (3.3) from $t - r$ to $t > t_1 + 3r$, we obtain

$$\log[Z(t)] - \log[Z(t - r)] + \sum_{i=1}^n \int_{t-r}^t H_i(s) k(s) ds = 0, \quad t > t_1 + 3r$$

Or equivalently,

$$\log[k(t)] = \sum_{i=1}^n \int_{t-r}^t H_i(s) k(s) ds = 0, \quad t > t_1 + 3r \quad (3.4)$$

Define

$$y = \liminf_{t \rightarrow \infty} k(t) \quad (3.5)$$

where $y \geq 1$. We have two possibilities: (i) y may be finite (ii) y may be infinite. Suppose y is finite,

then there exists a sequence $\{t_n\} \rightarrow \infty$ as $n \rightarrow \infty$ such that

$$\lim_{n \rightarrow \infty} k(t_n) = y \quad (3.6)$$

From (3.6) we have

$$\log[k(t)] = \sum_{i=1}^n \int_{t_n-r}^{t_n} H_i(s) k(s) ds = k(t_n) \sum_{i=1}^n \int_{t_n-r}^{t_n} H_i(s) ds \quad (3.7)$$

Where $t_n - r < t_n$, $n = 1, 2, 3, \dots$. Taking limit of both sides of (3.7) as $n \rightarrow \infty$ yields

$\text{Log}|y| \geq y \left(\liminf_{n \rightarrow \infty} \sum_{i=1}^n \int_{t_n-r}^{t_n} H_i(s) ds \right)$. That is,

$$\frac{\text{Log}|y|}{y} \geq \lim_{t \rightarrow \infty} \sum_{i=1}^n \int_{t-r}^t H_i(s) ds \quad (3.8)$$

Applying the fact that $\sup_{y \geq 1} \frac{\log|y|}{y} = \frac{1}{e}$ to (3.8) gives $\liminf_{t \rightarrow \infty} \sum_{i=1}^n \int_{t-r}^t H_i(s) ds \leq \frac{1}{e}$ which contradicts

the first part of the hypothesis of Lemma 1. By considering the second possibility, we contradict the other part of the Lemma. It must be that $Z(t)$ is almost certainly oscillatory. There certainly exists a subset $\Omega^* \subseteq \Omega$ such that

$$\Omega^* = \left\{ w \in \Omega : \liminf_{t \rightarrow \infty} \int_{t-\Gamma}^t \sum_{i=1}^n H_i(s) ds > \frac{1}{e} \text{ and } \liminf_{t \rightarrow \infty} \int_{t-\Gamma/s}^t \sum_{i=1}^n H_i(s) ds > 0 \right\} \quad (3.9)$$

with $P[\Omega^*] = 1$. Then as H_i and $f(t) = t - \Gamma$ satisfy the hypothesis of Lemma 1, it follows that the trajectory $Z(., w)$ is oscillatory and so the path $X(., w)$ is oscillatory and hence as the subset $\Omega^* \subseteq \Omega$ exists, it follows that the solution $X(t)$ of the SDDE (2.1) is almost surely oscillatory.

By property (2.4) of each $H_i(.,)$, we observe that

$$\begin{aligned} \int_t^{t+r_{ii}} H_i(s) ds &= \int_t^{t+r_i} -b \exp\left(-\left(a - \frac{\mu^2}{2}\right)r_i \exp(-\mu(B(s) - B(s-r_i)))\right) ds \\ &\geq -b \left(\max\left(1, \exp\left(-\left(a - \frac{\mu^2}{2}\right)r_i\right)\right) \int_{t-r_{ii}}^t \exp(-\mu(B(s) - B(s-r_i))) ds \right) \end{aligned}$$

It is observed (See Appleby and Buckwar [2005]) that the sure event $\Omega^* \subseteq \Omega$ as defined above exists eventually whenever

$$\lim_{t \rightarrow \infty} \sup \int_{t-r_{ii}}^t \exp(-\mu(B(s) - B(s-r_i))) ds = \infty. \quad (3.10)$$

and hence (2.1) has an oscillatory solution almost certainly.

Final Remark:

In the stochastic delay differential equation (2.1), under theorem 1, the important factor that stimulates oscillation is equation (3.10), which must always occur in the stochastic case as a result of the presence of the multiplicative noise. If the delays are small enough, the integral in (3.10) is made so small that the condition in Lemma 2 holds in the deterministic case (2.2) and at that instant, a non-oscillatory solution occurs in (2.2) but this cannot happen in the SDDE (2.1) as a result of (3.10). Hence the multiplicative noise sustains oscillation in the stochastic case (2.2) even when the non-stochastic equation (2.3) has a non-oscillatory solution. Therefore, the multiplicative noise stimulates oscillation about the zero equilibrium solution which may not necessarily be present in the deterministic case where $\mu = 0$. It should be noted that the noise has not entirely replaced the time delays as the cause of the oscillation. However, the time delay is no longer the sole cause of the oscillatory phenomenon of the stochastic delay differential equation.

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THEORETICAL ERROR ESTIMATE OF THE COMPLETELY DISCRETE SOLUTION OF LINEARIZED STOCHASTIC CAHN-HILLIARD EQUATION

Ignatius N. Njoseh

Department of Mathematics and Computer Science, Delta State University, Abraka

Abstract

We studied the finite element analysis of the linearized stochastic partial differential equation (linearized stochastic Cahn-Hilliard equation). The finite element method is used to discretize the equation. Based on the finite elements, the completely discrete approximation scheme was formulated by applying the backward Euler difference approximation in time. The completely discrete solution was interpreted in terms of analytic semigroup and converted to variation of constant formula using the rational functions definition to establish strong convergence rate for the completely discrete scheme.

1.0 Introduction

Stochastic Partial Differential Equations (SPDEs) are important tools in the modelling of complex phenomena and real life problems, such as, turbulence and pattern formation and to predict trends in the stock market or in weather, ground water flow, chemical reactions and heat emissions etc. They are also used for biological modelling and within the fields of medicine and engineering. Although the qualitative properties of these SPDEs have been thoroughly studied, the numerical approximations are imperative since the exact explicit solutions may not be readily available or obtained in usable form. This is because the numerical approximations of such equations have received less attention in recent research activities. Furthermore, the study of qualitative properties of SPDEs, involving the superprocess or simple variants of the heat equation, is also largely finished (cf: Mueller (2006)). Left to itself, SPDE might have become a dying field. Luckily, scientists are jumping into the field with a vengeance, and SPDE is expanding in all directions. We believe that the sciences will continue to provide important SPDE models and conjectures since scientists seem to have finally grasped the importance of SPDE models.

The purpose of this paper is to present numerical schemes and error estimates of the solution of linearized Stochastic Cahn-Hilliard

$$\begin{aligned} \dot{y} + A^2 y &= \dot{W}, \text{ in } \Omega \times [0, T], \quad y(0, \cdot) = y_0 \text{ in } \Omega \\ \frac{\partial y}{\partial n} &= \frac{\partial \Delta y}{\partial n} = 0 \text{ on } \partial \Omega \times [0, T] \end{aligned} \quad (1.1)$$

where $y(t)$ is a random process that takes values in $L_2 \Omega$, Ω is a bounded domain in \mathbf{R}^d ,

$d \leq 3$ with a sufficiently smooth boundary $\partial \Omega$. Δ is the Laplacian operator and W is a standard Brownian motion defined on a filtered probability space $(\Omega, F, \{F_t\}_{t \geq 0}, P)$.

Equation (1.1) is a fourth order heat equation used to model complicated phase separation and Coarsening phenomenon in a melted alloy that is quenched to a temperature at which only two different concentration phases can exist stably. This was developed by Cahn and Hilliard in 1958. (For more physical background on this equation, see Novich-Cohen and Segel (1984)). The existence and uniqueness of the solution of (1.1) has been a subject of study for a long time (cf Da Prato and Zabczyk (1992), Debussche and Zambotti (2006)). Finite element approximations of the deterministic form of (1.1) was analyzed in the L_2 -norms in Elliot and Larsson (1992), and Cardon-Weber (2000) studied the explicit and implicit discretization schemes of (1.1) in dimensions $d \leq 3$.

So much work has been done on the finite element analysis of the deterministic version of (1.1) but a little literature is available for the stochastic version. We shall now review some available literature.

Cardon-Weber (2000, 2001) established existence and uniqueness of a function-valued solution of the stochastic Cahn-Hilliard equation (1.1) in dimension $d \leq 3$. Here, the driving noise is the space-time white noise with non-linear diffusion coefficient. Under some non-degeneracy conditions on the diffusion coefficient, the author showed that the solution is locally differentiable in the sense of the Malliavin calculus and the law of the solution is absolutely continuous with respect to Lebesgue measure and obtained the explicit and implicit discretization schemes for (1.1). She observed that the polynomial growth of the drift term made her require the diffusion coefficient to be bounded, and proved convergence in probability (respectively in L_p with a given rate of a localized version) of the scheme, uniformly in space and time, that is, under some assumptions.

Yan (2003a) considered the finite element method for a stochastic parabolic partial differential equation of second order forced by additive space-time noise in the multi-dimensional case

$$dy + Aydt = dW, \quad \text{for } 0 < t \leq T, \quad \text{with } y(0) = y_0 \quad (1.2)$$

in the Hilbert space H with inner product (\cdot, \cdot) and norm $\|\cdot\|$, where $y(t)$ is a H -valued random process, A is a linear, self-adjoint, positive definite, not necessarily bounded operator with a compact inverse, densely defined in $D(A) \subset H$, where $\dot{W}(t)$ is a Wiener process defined on a probability space (Ω, F, P) and $u_0 \in H$. Yan set up a finite element analysis and applied the semigroup property generated by A to obtain optimal strong convergence estimates in the L_2 and H^{-1} norms with respect to spatial variable.

Also Njoseh and Ayoola (2008) discussed the finite element method for non linear for of (1.1) (i.e., with $f(y) = y^3 + y$) and obtained error estimates for both the semidiscrete and fully discrete solutions. Here the fully discrete scheme was obtained by applying the backward Euler time stepping finite difference method.

Hence, we shall be analyzing the error estimates of the solution of the linearized equation (1.1) using the finite element method. Our main aim here is to derive the semi-discrete and fully discrete schemes and obtain the error estimates using the analytic semigroup properties while following the methods adopted in recent works of Bin Li (2004), Elliot and Larsson (1992), Njoseh and Ayoola (2008), Yan (2003a and 2003b). The outline of this paper is as follows: In section 2, we present a few areas where (1.1) can be applied. In section 3, we explore the theoretical framework within which we will be working. We will particularly look at the Hilbert space. In section 4, we discuss our main result which is the semi-discrete and fully discrete error estimates for the problem under review.

2.0 Areas of application

Areas of application of equation (1.1) include the following

- i. Protein diffusion on charged membranes: Here lipids are allowed to move in the membrane plane according to a diffusion-like Cahn-Hilliard equation where segregation rates are in proportion to the Laplacian of their chemical potential. The local chemical potentials are derived from the free energy functional that depends on local lipid component densities and are calculated.
- ii. Chemical reactions and Calcium dynamics in cells.
- iii. Stochastic models in polymer field theory such as chemical mixtures and separations in polymer industries etc

3.0 Theoretical Framework

We now present definitions and notations that will be used throughout this work. Let

$H = L_2(\Omega)$ with inner product $(u, v) = \int_{\Omega} uv dx$ and corresponding norm $\|\cdot\| = (\cdot, \cdot)^{\frac{1}{2}}$. Let $A = -\Delta$

with domain $D(A) = H_0^1 \cap H^4$ where the spaces H^4 and H_0^1 are as defined below.

$$H^s = \{v \in L_2 : D^{\alpha} v \in L_2, |\alpha| \leq s\}$$

and

$$H_0^1 = \{v \in H^1 : v = 0 \text{ on } \Gamma = \partial\Omega\}$$

The space H^s has the inner product

$$(v, w)_s = \sum_{|\alpha| \leq s} \int_{\Omega} D^{\alpha} v D^{\alpha} w dx$$

and a corresponding norm $\|v\|_s = (v, v)_s^{\frac{1}{2}}$.

Define the space $H^s = H^s(\Omega) = D(A^{\frac{s}{2}})$, with norm $\|v\|_s = \|A^{\frac{s}{2}} v\|$ for any $s \in \mathbb{R}$.

Hence from Parseval's relation,

$$\|v\|_s^2 = \|A^{\frac{s}{2}} v\|^2 = \sum_{j=1}^{\infty} \lambda_j^s \hat{v}_j^2,$$

where λ_j are eigenvalues of A and $\hat{v}_j = (v, \phi_j)$ with ϕ_j an orthonormal basis of corresponding eigenfunctions.

For any Hilbert space, H , we define

$$L_2(\Omega; H) = \{v : E\|v\|_H^2 = \int_{\Omega} \|v(w)\|_H^2 dP(w) < \infty\}$$

with norm $\|v\|_{L_2(\Omega; H)} = (E\|v\|_H^2)^{\frac{1}{2}}$

Let HS denote the space of Hilbert – Schmidt operators from H to H , i.e.,

$$HS = \{\psi \in L(H) : \sum_{j=1}^{\infty} \|\psi \phi_j\|^2 < \infty\}$$

with norm

$$\|\psi\|_{HS} = \left(\sum_{j=1}^{\infty} \|\psi \phi_j\|^2 \right)^{\frac{1}{2}}$$

where $H = L_2$ and $\{\phi_j\}$ is an arbitrary orthonormal basis for H . Let E denote expectation and $\psi(s) \in HS$, then $\int_0^t \psi(s) dW(s)$ can be defined to have the isometry

$$E \left\| \int_0^t \psi(s) dW(s) \right\|^2 = \int_0^t \|\psi(s)\|_{HS}^2 ds$$

We assume that $W(t)$ is a Wiener process with covariance operator Q . This process may be considered in terms of its Fourier series. Suppose that Q has eigenvalues $\gamma_i > 0$ and corresponding eigenfunctions ξ_i . Then

$$W(t) = \sum_{i=1}^{\infty} \gamma_i^{\frac{1}{2}} \xi_i \beta_i(t)$$

where β_i , $i = 1, 2, \dots$ is a sequence of real-valued independent identically distributed Brownian motions.

4.0 Semi-discrete Scheme

Then with the definition of A and $D(A)$ we can write (1.1) as

$$y_t + A^2 y = dW, \quad t > 0, \quad y(0) = y_0 \quad (3.1)$$

having a mild solution of

$$y(t) = E(t)y_0 + \int_0^t E(t-s) dW(s).$$

Let S_h be a family of finite element spaces, where S_h consists of continuous piecewise polynomials of degree $r \leq 2$ with respect to the triangulation T_h of Ω . We shall also assume that $\{S_h\} \subset H_0^1(\Omega)$. According to the standard finite element method, the semi-discrete problem of (3.1) is to find $y_h(t) \in S_h$, such that

$$y_{h,t} + A_h^2 y_h = d\hat{W}, \quad t > 0, \quad y_h(0) = P_h y_0$$

where \hat{W} is the discrete approximate of W .

The mild solution is

$$\hat{y}_h(t) = E_h(t) P_h y_0 + \int_0^t P_h E_h(t-s) d\hat{W}(s)$$

where the operator $A_h : \dot{S}_h \rightarrow \dot{S}_h$ (the discrete Laplacian).

The error bound in the semi-discretization scheme is

Theorem 4.1

Let y_h be the spatially semi-discrete approximate solution of order r and with mesh parameter h , and let the initial approximation be chosen as the L_2 -projection of the exact initial value y_0 . Then if for $r \leq 2$ and $\|A^{(\gamma-1)/2}\|_{HS} < \infty$, for $\gamma \in [0, 4]$ we have

$$\|y_h(t) - y(t)\|_{L_2} \leq Ch^\gamma (\|y_0\|_{L_2(\Omega, \dot{H}^\gamma)} + \|A^{(\gamma-1)/2}\|_{HS}), \quad 0 < t \leq T$$

5.0 Fully Discrete Approximation

We now formulate the fully discrete approximation of (1.1) based on the backward Euler method in time. Here we replace the time derivative by a backward difference quotient

$\partial_t Y_n = \left(\frac{Y^n - Y^{n-1}}{k} \right)$ where k is the time step and Y^n is the approximation to y at time $t_n = nk$, $n = 1, 2, \dots$.

For equation (1.1), we pose the fully discrete approximation problem as follows: Find $y_h \in Y_h$; such that

$$y_{h,t} + A_h^2 y_h = \partial \tilde{W}, \quad t > 0, \quad y_h(0) = P_h y_0$$

and applying the implicit Euler method, for $k = \Delta t$, $t_n = nt$, $\Delta W^n = W(t_n) - W(t_{n-1})$ we have for $Y^n \in S_h$, $Y^0 = P_h y_0$;

$$\left(\frac{Y^n - Y^{n-1}}{k} \right) + A_h^2 Y^n = P_h \left(\frac{\tilde{W}(t_n) - \tilde{W}(t_{n-1}))}{k} \right), \quad t_n > 0 \quad (5.1)$$

$$\begin{aligned} Y^n - Y^{n-1} + k A_h^2 Y^n &= P_h \Delta \tilde{W}^n \\ Y^n - Y^{n-1} + k A_h^2 Y^n &= P_h (\tilde{W}(t_n) - \tilde{W}(t_{n-1})) \end{aligned} \quad (5.2)$$

and the variation of constants formula for

$$Y(t_n) = E(t_n) Y^0 - \int_0^{t_n} E(t_n - s) P_h dW(s)$$

becomes

$$\begin{aligned} Y^n &= E_{kh} Y^{n-1} - E_{kh} P_h \Delta W^n \\ Y^n &= E_{kh}^n P_h y_0 - \sum_{j=1}^n E_{kh}^{n-j+1} P_h \Delta W^j \end{aligned} \quad (5.3)$$

where $E_{kh} = (1 + k A_h^2)^{-1}$

5.1 Error Estimate

We now recall the following estimate from Yan (2003a) which we shall call a lemma.

Lemma 5.1

Let $B_n(t) = E_{kh}^n P_h - E(t_n)$, then for $0 \leq \gamma \leq 4$, $\|B_n v\| \leq C \left(k^{\frac{\gamma}{2}} + h^\gamma \right) |v|_\gamma$

and

$$\left(k \sum_{j=1}^n \|B_j v\|^2 \right)^{\frac{1}{2}} = \|B_n v\|_{L_2([0, T]; H)} \leq C \left(k^{\frac{\gamma}{2}} + h^\gamma \right) |v|_{\gamma-1}$$

where $|v|_\gamma = \|A^{\gamma/2} v\|$ for $\gamma \in \mathbf{R}$.

The error estimate for the fully discrete approximation is given below.

Theorem 5.1

Let y be the solution of (1.1), Y^n the solution of (5.2) and $e_n = Y^n - y(t_n)$. If $\|A^{(\gamma-1)/2}\|_{HS} < \infty$ for some $0 \leq \gamma \leq 4$, then

$$\|e_n\|_{L_2(\Omega, H)} = \|Y^n - y(t_n)\| \leq C(k^{\gamma/2} + h^\gamma) \left(\|y_0\|_{L_2(\Omega, \dot{H}^\gamma)} + \|A^{(\gamma-1)/2}\|_{HS} \right). \quad (5.4)$$

If $W(t)$ is a Wiener process with covariance operator $Q = I$, we have

$$\|e_n\|_{L_2(\Omega_H)} \leq C(k^{\gamma/2} + h^\gamma)(1 + \|y_0\|_{L_2(\Omega_H)}) \text{ for } 0 \leq \gamma \leq 2.$$

Proof:

$$\begin{aligned} \text{Since } e_n &= Y^n - y(t_n) \text{ and } B_n(t) = E_{kh}^n P_h - E(t_n) \text{ where} \\ Y^n &= E_{kh}^n P_h y_0 + \sum_{j=1}^n \int_{t_{j-1}}^{t_j} E_{kh}^{n-j+1} P_h d\widehat{W}(s) \text{ with } E_{kh}^n = r(kA_h^2)^n \text{ and} \\ y(t_n) &= E(t_n)y_0 + \int_0^{t_n} E(t_n-s)d\widehat{W}(s)ds, \end{aligned}$$

we have

$$\begin{aligned} e_n &= E_{kh}^n P_h y_0 + \sum_{j=1}^n \int_{t_{j-1}}^{t_j} E_{kh}^{n-j+1} P_h d\widehat{W}(s) - \left(E(t_n)y_0 + \int_0^{t_n} E(t_n-s)d\widehat{W}(s) \right) \\ &= \underbrace{E_{kh}^n P_h y_0 - E(t_n)y_0}_I + \underbrace{\sum_{j=1}^n \int_{t_{j-1}}^{t_j} B_{n-j+1} d\widehat{W}(s)}_{II} + \underbrace{\sum_{j=1}^n \int_{t_{j-1}}^{t_j} (E(t_n-t_{j-1}) - E(t_n-s))d\widehat{W}(s)}_{III} \end{aligned}$$

Thus

$$\|e_n\| \leq C(\|I\| + \|II\| + \|III\|)$$

For *I*, we have from Lemma 5.1 where $v = y_0$

$$\begin{aligned} \|I\| &= \|B_h v\| \leq C(k^{\gamma/2} + h^\gamma)|y_0|_\gamma \\ &\leq C(k^{\gamma/2} + h^\gamma)\|y_0\|_{L_2(\Omega_H)} \end{aligned}$$

For *II*, we have, by the isometry property

$$\begin{aligned} E\|II\|_{L_2(\Omega_H)}^2 &= E\left(\left\|\sum_{j=1}^n \int_{t_{j-1}}^{t_j} B_{n-j+1} d\widehat{W}(s)\right\|^2\right) \\ &\leq \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|B_{n-j+1}\|_{HS}^2 ds \\ &= \sum_{i=1}^\infty \left(k \sum_{j=1}^n \|B_{n-j+1} \varphi_i\|_{HS}^2\right) \\ &= k \sum_{i=1}^\infty \sum_{j=1}^n \|B_{n-j+1} \varphi_i\|_{HS}^2, \text{ where } \{\varphi_i\} \text{ is as in section (3.0).} \\ &\leq C \sum_{i=1}^\infty (k^{\gamma/2} + h^\gamma)^2 |\varphi_i|_{\gamma-1}^2 \\ &\leq C(k^\gamma + h^{2\gamma}) \sum_{i=1}^\infty |\varphi_i|_{\gamma-1}^2 \\ &= C(k^\gamma + h^{2\gamma}) \sum_{i=1}^\infty \|A^{(\gamma-1)/2} \varphi_i\|^2 \text{ by Parseval's relation} \\ &= C(k^\gamma + h^{2\gamma}) \|A^{(\gamma-1)/2}\|_{L_2}^2 \end{aligned}$$

For *III*, we have, by the isometry property again

$$\begin{aligned} E\|III\|_{L_2}^2 &= E\left(\left\|\sum_{j=1}^n \int_{t_{j-1}}^{t_j} (E(t_n-t_{j-1}) - E(t_n-s))d\widehat{W}(s)\right\|^2\right) \\ &\leq \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|E(t_n-t_{j-1}) - E(t_n-s)\|_{HS}^2 ds \\ &= \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|E(t_n-s)E(s-t_{j-1}) - I\|_{HS}^2 ds \\ &= \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|A^{\gamma/2} E(t_n-s) A^{-\gamma/2} (I - E(s-t_{j-1}))\|_{HS}^2 ds \\ &\leq \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|A^{\gamma/2} E(t_n-s)\|_{HS}^2 \|A^{-\gamma/2} (I - E(s-t_{j-1}))\|^2 ds \\ &= Ck^\gamma \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|A^{\gamma/2} E(t_n-s)\|_{HS}^2 ds \\ &\leq Ck^\gamma \|A^{(\gamma-1)/2}\|_{HS}^2 \sum_{j=1}^n \int_{t_{j-1}}^{t_j} \|A^{1/2} E(t_n-s)\|^2 ds \\ &\leq Ck^{2\gamma} \|A^{(\gamma-1)/2}\|^2 \end{aligned}$$

Hence

$$\|e_n\|_{L_2(\Omega, H)} \leq C(k^{\gamma/2} + h^\gamma)(\|y_0\|_{L_2(\Omega, \dot{H}^\gamma)} + \|A^{(\gamma-1)/2}\|_{HS}), \quad 0 < t \leq T$$

which concludes the proof.

6.0 Conclusion

The strong convergence rate in both the spatial and time steps can be computed. This can be done if the finite element solution computed on a very fine mesh is considered as the true solution and the finite element solutions computed on the less finer meshes are compared with this numerically obtained “true solution”. This is due to the fact that the true solution to the SPDE (1.2) itself is a random process and is not known explicitly.

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GENERATING FUNCTIONS APPROACH TO DETERMINATION OF THE SIGNIFICANCE OF GENE CLUSTERS.

J.N. Igabari

Dept of Mathematics and Computer Science, Delta State University, Abraka

Email: jn_igabari@yahoo.com

Abstract

There are many statistical tests to determine whether observed similarities in gene content are due to history or chance. Most of these methods are appropriate for whole genomic analyses. Researchers are gradually focusing on tests, for estimating significance when a single pair of genomic region is under consideration. Under the null hypothesis of random gene order, a method based on generating functions is proposed for estimating the significance of orthologous gene clusters. This proposed method can be used to determine the significance of gene clusters even in the absence of complete genomic content.

KEYWORDS

Generating functions, gene clusters, genome, window packing.

1 INTRODUCTION

A Genome, $G_i = (1, \dots, n_i)$ is modeled as an ordered set of n_i genes, ignoring chromosome break and physical distances between genes (Suzuki et al, 1989). It is assumed that genes do not overlap. Consider a simple model of two genomes, G_1 and G_2 , with identical gene content and one-to-one mapping between genes in G_1 and genes in G_2 . That is, every gene in G_1 has exactly one homolog in G_2 and vice versa. We define an orthologous cluster as a pair of windows, W_1 and W_2 , of length r_1 and r_2 selected from genomes G_1 and G_2 , respectively, that share m homologous gene pairs. In this simple model, the probability that pair of windows, of length r_1 and r_2 , have exactly m genes in common is simply the probability that m of the r_1 genes in W_1 also appear in W_2 and can be calculated using a hypergeometric distribution:

$$p_{1-1}(m) = \frac{\binom{r_1}{m} \binom{n_2 - r_1}{r_2 - m}}{\binom{n_2}{r_2}}$$

The probability that the windows share at least m genes is then $\sum_{i=m}^r p_{1-1}(i)$.

The 1-1 model requires a perfect, unambiguous homology mapping between G_1 and G_2 .

This may be possible after a recent speciation or polyploidization event. In general, however, because of variations in mutation rates, convergent evolution, non-homologous gene displacement and multi-domain proteins generated by exon shuffling, it may not be possible to identify a unique match. (Bochkina and Richardson, 2007). In that case, a many-to-many model is required. Genes are partitioned into families, such that any gene in a given family in G_1 can match any gene in the

same family in G_2 , the probability of finding a cluster by chance increases with family size. Consider, for example, the simple scenario where just one of the genes in w_1 matches f genes in G_2 . The probability of finding m matches for this one gene in a fixed size in G_2 increases since there are f possible matches for this one gene. However, it is surprisingly difficult to obtain a straightforward closed formula expressing this probability, even for this simple scenario. Therefore, accurate statistics requires a model of gene family size. However, this raises the challenge that once gene families are incorporated in the model, it is no longer easy to determine the expected number of matches in a window of size r .

2 GENERAL GENE FAMILY MODEL AND CLUSTER STATISTICS

The problem of identifying true homologs has been much debated and numerous solutions have been proposed {Durand and Sankoff (2003)}. The first step is typically sequence comparison. A variety of approaches are applied to rule out false positives or negatives due to weak sequence similarity and /or matches based on homologous domains in otherwise unrelated sequences. These include bi-directional best hits, imposing a minimum alignment length requirement and phylogeny reconstruction [Jarett and Ruggiero (2008)]. Despite these efforts, homology frequently remains unresolved. Furthermore, gene duplications that occur after the specification separating G_1 and G_2 , result in situations where a gene in one genome has two or more legitimate orthologs in the other [Cole et al, 2007].

We therefore extend this model to include gene families. A gene family is a set of *homologous genes*; that is, genes that share a common ancestor, through either duplication (paralogs) or specification (orthologs) [Russel (1986)]. Gene family membership in our model does not depend on inherent functional or structural properties of the family but rather on what type of information the user brings to bear on identification of homologous relationships. We define a gene family to be the set of *indistinguishable* homologous genes; i.e., homologous genes, where sub-family classification cannot be further disambiguated.

We will assume that the set of genes in genomes G_1 and G_2 can be partitioned into non-intersecting gene families. Let $f_{ij} \subset G_i$ denote the members of the j th gene family in genome i . Then, the j th gene family, $f_j = f_{1j} \cup f_{2j}$, is a set of genes such that each gene in f_i is homologous to all other genes in f_j and only those genes. There are $\phi_{ij} = |f_{ij}|$ genes in the j th family in genome G_i . Let $\mathcal{F} = \{f_j\}$ be set of all gene families in both genomes. In the gene family model, we define an orthologous gene cluster to be a pair of windows of length r_1 and r_2 , drawn from G_1 and G_2 , respectively, that have m gene families in common.

We consider a test for an individual cluster based on the probability of observing a cluster in two genomes with uniform random gene order (a “random genome”). In calculating cluster probabilities for the general case, we will need to count the number of ways that a window of a particular size can be filled with a given set of gene families in several contexts. We therefore derive a general solution to this problem using generating functions, and combinatorial analysis which can be used to determine a sequence of interest from the coefficients of a power series [Durand and Sankoff (2003)]. Here the sequence of interest is the number of ways filling the window. It is this formalism that allows us to compute cluster probabilities efficiently.

3 METHODOLOGY

a Window packing and the generating function formulation

Define \mathcal{T} to be set of λ gene families of arbitrary size $\phi_1, \dots, \phi_\lambda$. Given the sample space of all sets of w genes sampled from genome of size n , we wish to enumerate those that contain at least one gene from each family in \mathcal{T} . Since we do not take into account the order of genes in a window, this enumeration is equivalent to finding all window packings. The generating function formulation

allows us to determine the number of such window packings, denoted by $\mathcal{N}(w, \lambda, \mathcal{T})$. We represent contribution of the j th family in \mathcal{T} by the generating function

$$\alpha_j(t) = \binom{\phi_j}{1}t + \binom{\phi_j}{2}t^2 + \dots + \binom{\phi_j}{\phi_j}t^{\phi_j}. \quad (1)$$

The coefficient of t^i in $\alpha(t)$, denoted by $[t^i]\alpha_j(t)$, represents the number of ways of choosing i genes from j th family. The contributions of all λ families to the window can then be derived from the product of their generating functions:

$$\alpha(t) = \prod_{j=1}^{\lambda} \left[\binom{\phi_j}{1}t + \binom{\phi_j}{2}t^2 + \dots + \binom{\phi_j}{\phi_j}t^{\phi_j} \right]. \quad (2)$$

The coefficient $[t^w]\alpha(t)$ gives the number of ways of filling w slots with genes from the λ families, which is just $\mathcal{N}(w, \lambda, \mathcal{T})$. Note that the t^w term in $\alpha(t)$ will be sum of products of the form $\beta_1 t^{x_1} \cdot \beta_2 t^{x_2} \dots \beta_{\lambda} t^{x_{\lambda}} = \left(\prod_j \beta_j \right) t^w$, where the exponents of the dummy variable, t , sum to w . by inspecting Equation (2), we see that since β_j is the coefficient of t^{x_j} , it must be of the form $\beta_j = \binom{\phi_j}{x_j}$. The term $[t^w]\alpha(t)$ corresponds to packings containing x_1 genes from the first family, x_2 genes from the second family and so forth, where β_j corresponds to the number of ways of choosing x_j genes from j th gene family. Summing over all packings, we obtain

$$\mathcal{N}(w, \lambda, \mathcal{T}) = \sum_{(x_1, \dots, x_{\lambda})} \binom{\phi_1}{x_1} \binom{\phi_2}{x_2} \dots \binom{\phi_{\lambda}}{x_{\lambda}}, \quad (3)$$

where the sum is over the set of all λ -tuples $(x_1, \dots, x_{\lambda})$ such that

$$\sum_{j=1}^{\lambda} x_j = w, \quad \text{and } 1 \leq x_j \leq \phi_j, \forall j. \quad (4)$$

Let us illustrate the window packing problem with a simple example. Suppose we wish to find the number of ways a window of size $w = 7$ can be packed with four gene families ($\lambda = 4$), such that the window has at least one gene from each gene family. Let the gene family size of \mathcal{T} be $\phi_1 = 1, \phi_2 = 2, \phi_3 = 3$ and $\phi_4 = 4$ and the 4-tuple (x_1, x_2, x_3, x_4) refers to a window packing that has x_1 genes from the first gene family, x_2 genes from the second gene family, x_3 genes from the third gene family and x_4 genes from the fourth gene family. In order to find all possible packings, we need to find the all 4-tuples satisfying equation (4); in this example $\sum_{j=1}^4 x_j = 7$. Since j th gene family can contribute x_j genes in $\binom{\phi_j}{x_j}$ ways, the 4-tuple (x_1, x_2, x_3, x_4) can contribute $\binom{\phi_1}{x_1} \binom{\phi_2}{x_2} \binom{\phi_3}{x_3} \binom{\phi_4}{x_4}$ window packings. For example, the tuple $(1, 1, 1, 4)$ can contribute $\binom{1}{1} \binom{2}{1} \binom{3}{1} \binom{4}{4} = 6$ window packings. Table 1 lists the set of all possible 4-tuples and the number of packings associated with each 4-tuple. By adding the number of packings for each 4-tuple, we get the total number of ways the window can be filled with genes from the four gene families as given in Equation (3). Here, $\mathcal{N}(7, 4, \mathcal{T}) = 76$.

λ - tuple	Number of
(x_1, x_2, x_3, x_4)	Packings
(1,1,1,4)	6
(1,1,2,3)	24
(1,1,3,2)	12
(1,2,1,3)	12
(1,2,3,1)	4
(1,2,2,2)	18
$\mathcal{N}(7,4,\mathcal{T})$	76

Table 1. Number of ways of packing a window of size $w = 7$ with 4-gene families of size $\{1,2,3,4\}$.

b Orthologous cluster with arbitrary gene families

We estimate the significance of gene cluster using the probability that two windows, arbitrarily chosen from two random genomes, share at least m gene families. We enumerate over all sets to k gene families, for each value of k from m to r . For each such set F , we determine the probability that w_1 contains only genes in families in F , including at least one from each family, followed by the conditional probability that at least l of the families in F also appear in W_2 .

Expressed formally, the probability that W_1 and W_2 share at least m gene families is.

$$q_o(m) = \sum_{k=m}^r \left[\sum_{\mathcal{F} \in \mathcal{F}^k} p^1(\mathcal{F}) \sum_{l=m}^k \sum_{\substack{E \in \mathcal{F}^l \\ E \subseteq \mathcal{F}}} p^2(E) \right], \quad (5)$$

where \mathcal{F} is the set of gene families in G_1 and G_2 . The probability that a given set, \mathcal{F} , of k gene families is seen in W_1 is

$$p^1(F) = \binom{n_1}{r_1}^{-1} \mathcal{N}(r_1, k, F) \quad (6)$$

where $\mathcal{N}(r, k, F)$ is the number of window packing given by equation (3). To determine, $p^2(E)$, we enumerate over all subsets of F of size l , where l ranges from m to k . For each subset E , we seek the probability that each family in E is represented in W_2 at least once and that no other family in F appears in W_2 . We exclude all other families in F to avoid over counting.

At least l slots in W_2 must be filled with genes in E . The remaining $r^2 - l$ slots may be filled either from families in E or from families that do not appear in W_1 ; i.e., genes from $\mathcal{F} \setminus F$. Let

z be the number of slots filled with genes from $\mathcal{F} \setminus F$. By considering all possible values of z , we obtain $p^2(E) = \binom{n_2}{r_2}^{-1} \sum_z \mathcal{N}(r_2 - z, l, E) \binom{n_2 - \phi(F)}{z}$ (7)

where $\phi(F) = \sum_{j \in F} \phi_{2j}$. The parameter z ranges from $\max\{0, r_2 - \phi(E)\}$ to $r_2 - l$ where $\phi(E)$ is defined as above. The first term in the numerator is the number of ways of filling $r_2 - z$ slots with genes from l families in E . The second term corresponds to all the ways of choosing the z outsiders from the set of genes not included in any gene family in W_1 . By substituting the expression in equation (3) in equations (6) and (7), we get a statistic for individual clusters in terms of n_1, r_1, n_2, r_2, m and the set of gene families in G_1 and G_2 . However, calculating this probability requires the enumeration of all subsets of k gene families. For each subset, we must enumerate all packings satisfying equation (4) and calculate a product of binomials for each packing. Computing this probability is prohibitively slow.

c Orthologous clusters with fixed size families

The complexity of calculating $q(m)$ can be substantially reduced under the assumption that all gene families are of equal size, ϕ . When gene families are of equal size, it is not necessary to enumerate \mathcal{F}^k , since all subsets of k gene families are indistinguishable. We can simply replace the first term, $\sum_F p^1(F)$, in equation (5) with the product of number of sets of k gene families times $p^1(k)$, the probability that exactly k gene families of size ϕ are represented in the window: $\sum_{F \in \mathcal{F}^k} p^1(F) = \binom{|\mathcal{F}|}{k} p^1(k)$. (8)

Invoking a similar transformation of the second term in equation (5), the probability that W_1 and W_2 share at least m gene families simplifies to

$$q_o(m) = \sum_{k=m}^r \left[\binom{n_f}{k} p^1(k) \sum_{l=m}^k \binom{k}{l} p^2(l) \right] \quad (9)$$

Under the fixed size assumption, $p^1(k)$ and $p^2(l)$ correspond to the probability that exactly k families appear in W_1 and exactly l families appear in W_2 , respectively. To calculate $p^1(k)$ and $p^2(l)$, we require an expression for $\mathcal{N}'(w, \lambda, \phi)$, the number of window packings when all families are of fixed size. When $\phi_j = \phi, \beta_j$ reduces to $\binom{\phi}{x_j}$ and equation (2) becomes

$$\alpha'(t) = \left[\binom{\phi}{1} t + \binom{\phi}{2} t^2 + \dots + \binom{\phi}{\phi} t^\phi \right]^\lambda \quad (10)$$

The number of ways observing λ gene families in a window of size w is given by $[t^w] \alpha'(t)$, yielding

$$\mathcal{N}'(w, \lambda, \phi) = \sum_{(x_1, \dots, x_\lambda)} \binom{\phi}{x_1} \binom{\phi}{x_2} \dots \binom{\phi}{x_\lambda} \quad (11)$$

where the sum is over the set of all λ -tuples (x_1, \dots, x_λ) satisfying Equation (4), under the constraint that $0 < x_1 \leq \phi, \forall i$. In this case, we can avoid enumerating the λ -tuples using the

following simplification. Note that the right hand side of equation (10) is a binomial series of the form $[(1+t)^\phi - 1]^\lambda$. By applying two binomial expressions, we obtain

$$\alpha'(t) = (-1)^\lambda \sum_{i=0}^{\lambda} (-1)^i \binom{\lambda}{i} \sum_{j=0}^{i*\phi} \binom{i*\phi}{j} t^j. \quad (12)$$

The number of ways of filling \mathbf{w} slots with genes from the λ fixed size families is just $[t^{\mathbf{w}}]\alpha'(t)$, yielding

$$\mathcal{N}'(\mathbf{w}, \lambda, \phi) = (-1)^\lambda \sum_{i=0}^{\lambda} \left[(-1)^i \binom{\lambda}{i} \binom{i*\phi}{\mathbf{w}} \right]. \quad (13)$$

Notice that the least $\left\lceil \frac{\mathbf{w}}{\phi} \right\rceil$ gene families are required to fill a window of size \mathbf{w} . Substituting the expression for $\mathcal{N}'(\mathbf{w}, \lambda, \phi)$ in equation (6) and restricting the lower bound on the dummy variable i to $\left\lceil \frac{r_1}{\phi} \right\rceil$, we obtain

$$p^1(k) = \binom{n_1}{r_1}^{-1} (-1)^k \sum_{i=\left\lceil \frac{r_1}{\phi} \right\rceil}^k \left[(-1)^i \binom{k}{i} \binom{i*\phi}{j} \right] \quad (14)$$

Similarly, $p^2(l)$, the probability that W_2 contains exactly l gene families is

$$\binom{n_1}{r_1}^{-1} \sum_z (-1)^l \sum_{i=\left\lceil \frac{r_2-z}{\phi} \right\rceil}^l \left[(-1)^i \binom{l}{i} \binom{i*\phi}{r_2-z} \binom{n_2-k\phi}{z} \right] \quad (15)$$

where z ranges from $\max\{0, -k\phi\}$ to $r_2 - l$.

4 DISCUSSION

The fixed size approximated and the use of generating functions to estimate window packings result in an efficient approximation to the probability that two windows, arbitrarily chosen from two random genomes, share at least \mathbf{m} gene families. The general gene family model requires implementation of an algorithm to enumerate all λ -tuples satisfying equation (4). Furthermore, it is necessary to compute the product of λ binomial terms for each of the tuples in the enumeration. In contrast, equations (14) and (15) require only a simple summation and can be mathematically. We can compute equation (9) using the number of genes in each genome, the window sizes, gene family sizes and the number of gene families shared between the window. We only need information about the local regions and the aggregate properties of the genomes to determine significance of individual clusters. We have, thus, presented a modified approach to determine the significance of individual gene clusters that takes gene family size into account. It can be used to determine the significance of gene clusters in the absence of complete genomic context. We estimate the significance of gene cluster by determining the probability that two regions, containing r_1 and r_2 genes respectively, share at least \mathbf{m} gene families. By using generating functions, we have developed practical expressions for the estimating probability of observing orthologous gene clusters in two genomes.

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THE EFFECT OF ALLIUM SATIVA (GARLIC) ON THE GROWTH OF FUNGI ISOLATED FROM LEATHER SHOES

O.O. Akpomie, N. J. Ehiwario, and C.A. Ozochi

Department of Microbiology, Faculty of Science, Delta State University Abraka,

Abstract

Mucor mucedo, *Aspergillus niger*, *Rhizopus nigricans*, *Penicillium chrysogenum* and yeast were isolated from men's leather shoes in Abraka. The phytochemical analysis showed the presence of flavonoids, alkaloids, tannins and saponins. The water extract was able to inhibit the growth of *M. mucedo*, *R. nigricans*, *P. chrysogenum* and yeast at a concentration of 0.375 g/ml while *A. niger* was inhibited at a minimum concentration of 0.500 g/ml. The zones of inhibition for *M. mucedo*, *A. niger*, *R. nigricans*, *P. chrysogenum* and the yeast were 4.20, 3.30, 4.10, 5.90 and 5.00 respectively. The physical property of the treated leather samples compared favorably well with that of untreated ones.

Key words: *Allium sativa*, leather, fungi, MIC.

INTRODUCTION

In the leather industry, efforts are being made to discover factors that affect the quality and durability of leather and leather products. Emphasis has been placed on the impact of mechanical tear and wear of shoes to the neglect of other contributing factors that could equally affect the quality of leather and leather products. Munson and Bode (1995) found that deterioration of shoes is caused mostly by water. Water dissolves tannins and other water-solubles which favour the growth of fungi and bacteria in the upper materials of leather and synthetic shoes (Akpomie et al., 2006). These microorganisms exhibit different hydrolytic tendencies which eventually result in deterioration.

Aminoquinolones and other chemicals have been used as a desiccant and as preservatives in leather and leather products against fungi but most of these chemicals have their attendant problems (Perumal et al., 1999). Some cause discoloration, others damage the leather fibres' matrix and some can cause environmental pollution. A lot of researches have been done on the use of plant extracts as antimicrobial but little has been done on the effectiveness of the plant extracts against fungi posing problems in leather and footwear.

There is therefore, the need to look into alternative preservatives, of little or no environmental hazards that will cause no damage to the leather and its products.

Garlic has been found to contain allicin, ajoene, flavonoids which are antimicrobial agents (Ankri & Mirelman, 2001). This study thus aims at investigating the activity of *Allium sativa* (garlic) extract (a natural product that will constitute very little or no environmental hazard) in preventing the growth of fungi in men's leather shoes.

Experimental preparation of extract

Peeled garlic cloves were properly washed with distilled water and ground in a mortar. The ground material was pressed and filtered through a piece of muslin cloth. The filtrate was collected in a sterile dark bottle and kept in the refrigerator at 40°C for further use (Pyun and Shin, 2005).

Phytochemical analysis of extract

The extract was tested for the presence of flavonoids, alkaloids, tannins and saponnins using the methods of Trease and Evans (1996).

Isolation and characterization of fungi

Sterile plates of potato dextrose agar (PDA) were inoculated with inoculum collected from sterile men's leather shoes. Sterile swab sticks with cotton wool partly wet with sterile peptone water were used aseptically to pick the inoculum. The plates were incubated at 28°C for 72h. Characterization of fungal isolates was done using the illustrations of Barnett and Hunter (1972).

Determination of minimum inhibitory concentration

The method of Niinez et al., 1998 was adopted using the following concentrations of the extract, 0.375 g/ml, 0.5 g/ml, 0.75 g/ml and 1.5 g/ml.

Determination of the effect of the extract on some physical properties of leather

Five pieces of leather samples were cut, soaked in 0.375 g/ml and 0.500 g/ml concentrations of the extract, one piece was not soaked in the extract (control). They were shaken in an orbit-environ shaker 18 at 28°C for 48 h. This was allowed to dry and left in the laboratory for three months for visual observation of bacterial growth. The shrinkage temperature and tensile strength of the leather samples were determined according to the Official Methods of SLTC (1999).

RESULTS AND DISCUSSION

The organisms identified were identical for most of the leathers shoe samples but the frequency of occurrence of each isolate differed. *Mucor and Aspergillus spp.* were more with an average frequency of 15 and 12 respectively. Yeast occurrence was least of all the isolates (Figure 1).

A total of 45 fungal isolates were gotten from the samples. *Mucor* sp. occurred most (33%) while yeast had the least occurrence (6.7%). The leather shoes wet from perspiration contained salt in addition to the materials in the shoe. This provided nutrient for microorganisms which eventually attacked the leather (Pyun and Shin, 2005). The fungi survived the effect of the polish and other leather treatments which is evidenced in the study of Raul et al. (1996), that some microorganisms are able to survive extreme conditions and treatment. The fungi were able to secrete enzymes which split and decompose the organic matters present in the shoes (Van and Botha, 1997).

Penicillium had the highest diameter of clear zone (5.9 cm) while *Aspergillus* had the least of zone inhibition (3.3 cm) as shown in Table 1. There were good inhibitory effects evidenced in the clear zones of inhibition. The concentration at which garlic extract inhibited the growth of *Rhizopus*, *Aspergillus*, *Penicillium* and Yeast was 0.375 g/ml while *Mucor* was inhibited at 0.50 g/ml. The extract had a good antimicrobial activity against all the fungal isolates (Table 2). Garlic contains compounds that react with thiol groups of various enzymes like alcohol dehydrogenase, RNA polymerase which can affect essential metabolism thus inhibiting the growth of the microorganisms (Bghalycin et al., 2006).

The result of the determination of shrinkage temperature and tensile strength of the leather (Tables 3 and 4) compared favorably well with that of the control. This suggests that the extract did not reduce the quality and durability of the leather.

CONCLUSION

R.nigricans, *M. mucedo*, *A. niger* and Yeast were isolated from the leather shoes. Uncontrolled growth of the fungi on leather footwear has diverse effects on the footwear and the wearer. The extract exhibited a high antimicrobial activity against all the isolates. In humans, fungi have been

observed to cause diseases such as athlete's foot. Growth of fungi on leather shoes may be combated with garlic extract being incorporated at some stages in leather or in the production of polish and other materials formulated to increase the quality and durability of leather.

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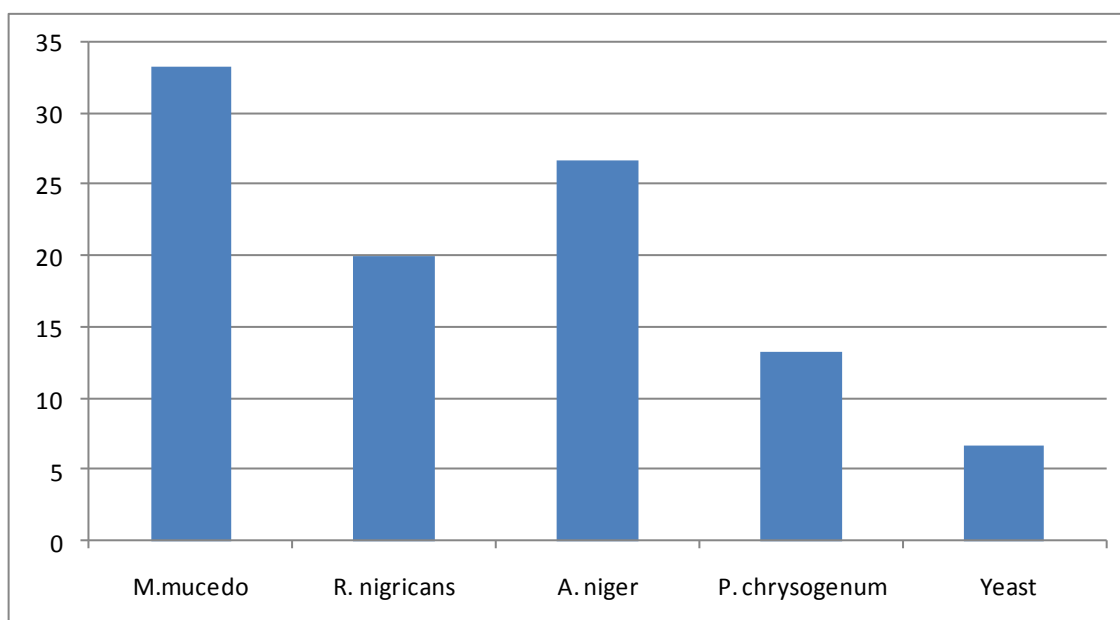


Figure 1 (colour online). Frequency of occurrence of the fungal isolates (%)
Y-axis -% frequency of occurrence of each isolate of a total of 45 isolates
X-axis - Fungal isolate

Table 1. Diameter (mm) of Zones of Inhibition of Garlic extract on the growth of fungal isolates.

Isolates	Zones of inhibition (mm)
<i>Mucor mucedo</i>	4.20
<i>Aspergillus niger</i>	3.30
<i>Rhizopus nigricans</i>	4.10
<i>Penicillium chrysogenum</i>	5.90
Yeast	5.00

Table 2. Evaluation of the minimum inhibitory concentration (MIC) of the extract on the isolates.

Isolates	MIC (g/ml)
<i>Mucor mucedo</i>	0.375
<i>Aspergillus niger</i>	0.500
<i>Rhizopus nigricans</i>	0.375
<i>Penicillium chrysogenum</i>	0.375
Yeast	0.375

EFFECTS OF DETERGENTS AND LOCAL SOAP “SODA” ON GERMINATION OF THE SEEDS OF MONOCOTS AND DICOTS

Erhenhi, A.H

Department of Botany, Delta State University, Abraka, Delta state
E-mail: mac_harrison7@yahoo.com

ABSTRACT

The healthy seeds of four crops, two monocots (*Sorghum bicolor* and *Zea mays*) and dicots (*Glycine max* and *Phaseolus vulgaris*) were treated with different detergents and local soap “ Soda” concentrations to ascertain the effects on their germination. Germination was hindered in the monocots and dicots treated with 5%, 10%, 15% and 20% of detergents (BONUX, ARIEL and OMO) whereas, germination occurred in plants treated with 5%, 10%, 15% and 20% concentrations of local soap “ Soda”. As the concentration of local soap increased from 5%-20%, the number of seeds per bag that germinated reduced. Detergents completely hindered the seed germination of dicots with an exception to monocots at low concentration which died off as time progresses. These results suggest that local soap waste water could be used for watering of vegetable garden while that from detergent cannot.

Key words: Germination, monocots, dicots

INTRODUCTION

The seed is the starting point in the life cycle of plants. A seed refers to the embryo, or immature plant, that will grow and develop into the seedling and ultimately into the matured plant. The seed coat (or testa) surrounds the seed and carries the nutritive source for germinating seedling. The seeds of plants may either be monocots (grasses, maize etc.) or dicots (legumes and peas)

Germination of seeds begins with the absorption of water which causes the seeds to swell and burst the testa if the testa is not permable (Eze and Ahonsi 1993). Also the first true growth occurs in the radicle, the tip of which emerges through the broken testa in the region of the micropyle and grows downwards into the soil. The growth of the embryo into a seedling is termed germination. Therefore, germination can be defined as the reawakening of a dormant embryo to active growth. This is often followed by an overall increase in metabolic activity. A seed that has not germinated because it is lacking one or more of the necessary requirements for germination is termed “quiescent”. Such seeds are simply “resting”, waiting for the appropriate condition for germination, when such seeds are given water, oxygen and a suitable temperature, a quiescent seed will germinate. However, when a seed is exposed to the appropriate conditions and failed to germinate, such a seed is described as “dormant”.

According to Carl (1998) detergents which contain high phosphate were toxic to the seedling of *zea mays* and thus have adverse effects on the germination of the seeds; and thus interfere with the natural growth processes of the seedlings. Rapid imbibition of water is the central activity that triggers off seeds germination. Once imbibed, water activates enzyme which were until then dormant in the seed (Berrie *et al.*, 1987). Though water is very important in germination, they showed that

seeds failed to germinate in water-logged soils. The failure to germinate is due to lack of oxygen in the soil because water would have displaced the soil air. Many seeds native to temperate climate must be stratified. Some seeds have a hard seed coat that needs to be naked (termed scarification) for germination. Still, other seeds require an exposure to heat in order to germinate (Bidwell, 1979).

It is generally recognized that germination of seeds depends upon both external and internal factors. Noggle and Fritze (1976) reported that germination may be blocked by the absence of some external factors. These factors could be absence of water, suitable temperature, sunlight etc. some seeds may be placed under suitable condition and still not germinate because of internal factors which include their genetic and physiology.

Germination and seedling establishment are quite essential for increase in population, and avoidance of extinction by a plant species. Triplett and Tessar (1960), enumerated the problems of seed germination which have not been thoroughly explored. The factor most commonly associated with failure of seedling to emerge are insufficient soil moisture, improper depth coverage, temperature, light and Humidity

Low concentration of phosphate in soap act as a fertilizer for seeds, and speed up the rate at which the seeds will germinate, (Mcbain and salmon 1970). Seeds usually will require initial high amounts of phosphorus, thus boost its germination. However, in later stages when the seedling has become established, additional phosphate is not needed, and will have no effect, rather have adverse effects. Detergent dissolves cell membrane and can denature proteins in seeds, which include enzymes. Since step 1 of germination is cutting through the hull of the seed with enzymes (Pemadasa and Lovelli, 1974). Detergent does slow down the germination processes in contrast to soap which speed up the breaking of seed dormancy.

MATERIALS AND METHODS

The seeds of *Phaseolus vulgaris*, *Glycine max*, *Sorghum bicolor* and *Zea mays* were obtained from Abraka main market and transported to Botany laboratory, Delta State University Abraka, identified by the Taxonomist in the department, labeled and kept in the herbarium. The detergents (Ariel, Bunux, and Omo) and local soap “Soda” were also purchased from Abraka main market labeled respectively as A,B,C and D. Electrical weigh balance (AR2140) was used to weigh samples of detergents, local soap and loamy soil. 50g, 100g, 150g and 200g of detergents and local soap were dissolved into 1000ml of distilled water to obtain the needed concentrations. Loamy soil and black polythene bags used for this study were obtained from site II of DELSU. Four concentrations 5%, 10%, 15% and 20% of the three detergents and local soap were prepared. One hundred grams (100g) each of the loamy soil were treated with the different concentrations of the detergents and local soap. Three seeds were planted per bag. Viability test was carried out by using the floating method. Different seeds of various plants were immersed in different containers of water and those that floated were regarded as being nonviable and discarded. As far as possible, viable seeds were selected for uniformity of size as this is important in assessing results obtained (Black, 1972)

RESULTS

The results on the figures (1-4) showed the effects of detergents and local soap on the germination of the seeds of monocots and dicots treated with varying levels of concentrations.

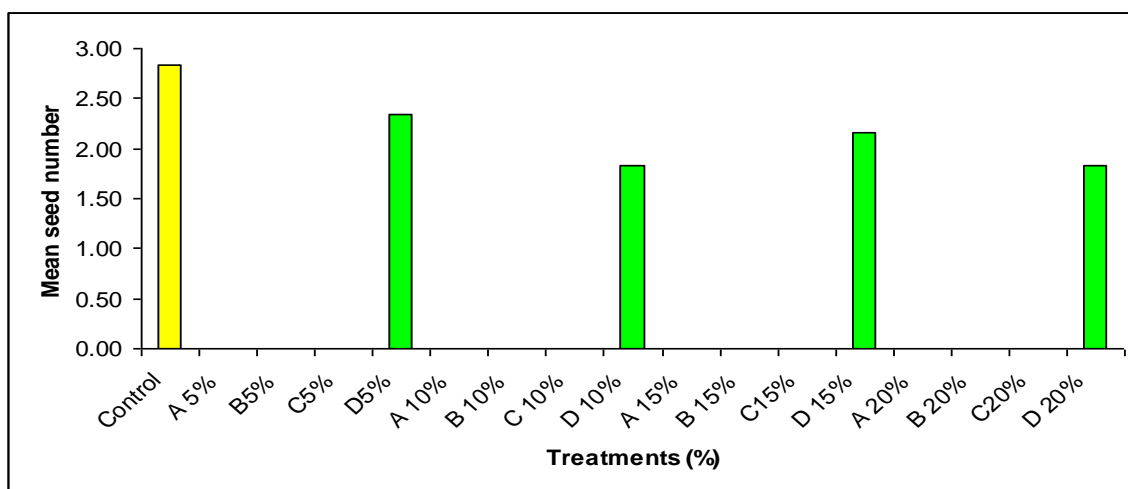


Figure 1 (colour online): Effects of Detergents and local soap on *Glycine max* seeds germination (A= Ariel, B= BUNUX, C= OMO, D= SODA)

The results of effects of the detergents and local soap show that, at day 1, the seeds of *Glycine max* treated with 5%, 10% and 15%, and 20% concentrations of local soap and control germinated. However, no germination of seeds was recorded in any of the treatments of concentrations of the three detergents (Figure 1). As at day 6, while all the seeds treated with 5%, 10% and 15%, 20% concentrations of local soap and control germinated i.e. hundred percent, no germination was obtained with the seeds treated with different concentrations of the three detergents.

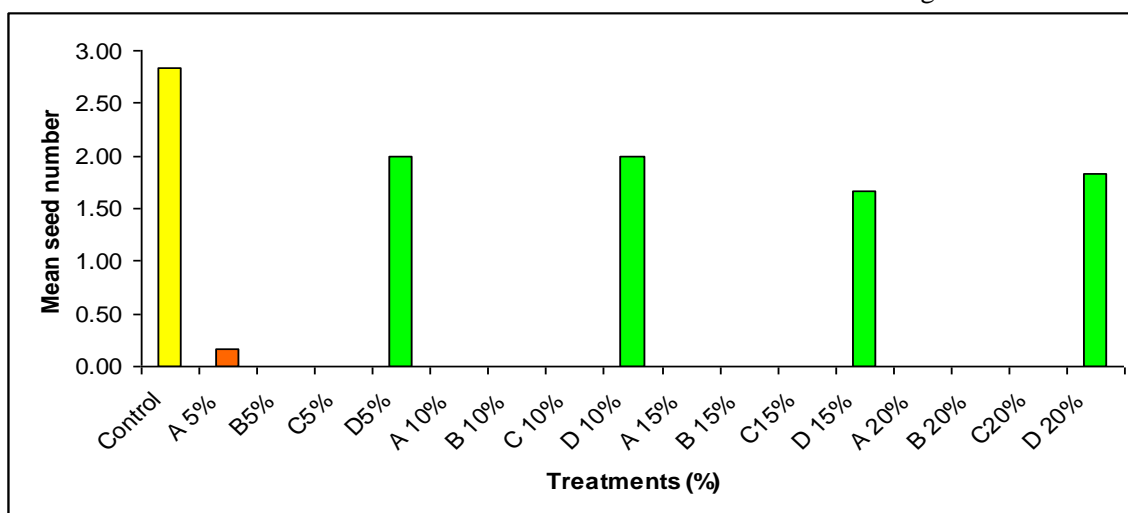


Figure 2 (colour online): Effects of Detergents and local soap on *Phaseolus vulgaris* seeds germination (A= Ariel, B= BUNUX, C= OMO, D= SODA)

The results presented in figure 2 shows two seeds of *Phaseolus vulgaris* germinated and one each germinated with seeds treated with 5%, 10% and 15% and 20% concentrations of local soap in day 1, there was no germination of seeds recorded with those treated with the detergents. In day 4, it was observed the seeds treated with Ariel detergent had one of the seeds germinated while those in control and 5%, 10% and 20% concentration had had 100% germination i.e. the three seeds planted at the bags germinated. However, at day 6, the one seed observed to have germinated in 5% Ariel detergent

treatment died off. Seeds treated with the three detergents were hindered from germination unlike what was recorded in control and all concentrations of local soap.

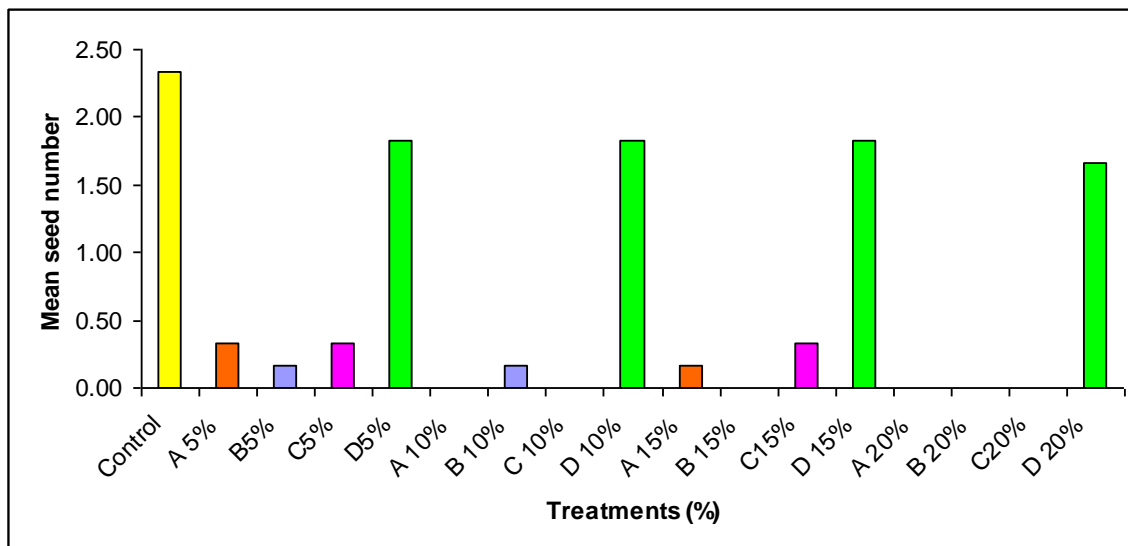


Figure 3 (colour online): Effects of Detergents and local soap on *Sorghum bicolor* seeds germination (A= Ariel, B= BUNUX, C= OMO, D= SODA)

Unlike, what was observed in the seeds of the two dicots above, germination was observed in three detergents. seeds of *Sorghum bicolor* treated with 5% Omo detergent concentration had one seed growth at day 2, 10% Bunux detergent concentration had one seed growth at day 4, 15% Omo detergent concentration had one seed growth at day 4. At day 6, the seeds planted in control and 5%, 10%, 15% and 20% concentrations of local soap recorded 100% germination i.e. the three seeds planted per bag germinated, whereas, the single seed that germinated in the two detergents died off (Figure 3)

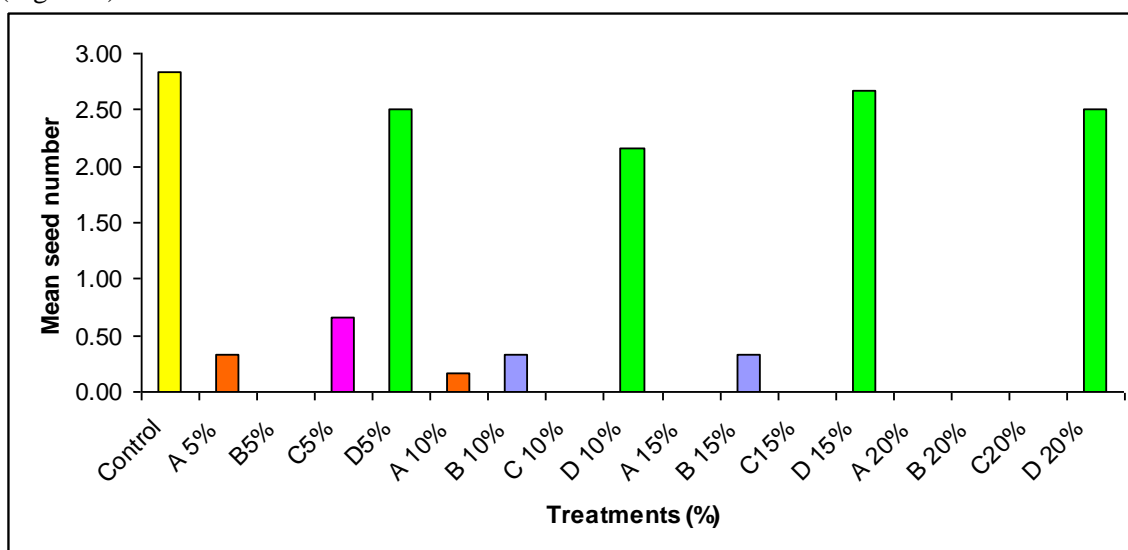


Figure 4 (colour online): Effects of Detergents and local soap on *Zea mays* seeds germination (A= Ariel, B= BUNUX, C= OMO, D= SODA)

The result obtained with the seeds of *Zea mays* (Figure 4) had a remarkable similarity with that obtained in figure 3 with the seeds of *Sorghum bicolor* (a monocot). There was germination recorded for seeds treated with some detergents concentrations. Germination was observed with seeds treated with 5% concentration of Ariel detergent at day 3, 5% concentration of Omo detergent at day 4, 10% concentration of Bunux at day 4 and no germination was recorded for seeds treated with 20% concentration of detergents (Figure 4). At day 6, the seeds planted in control and all concentrations of local soap recorded 100% germination i.e. the three seeds planted per bag germinated, whereas, the single seed that germinated in the two detergents died off (Figure 4).

DICUSSION

The adverse effects of detergents on the seed germination of monocots and dicots observed in Figures 1-4 may be solely due to the surfactant content of the detergents which tends to have a negative influence on seeds germination. This agrees with previous work of Cameron (1993), that detergent generally contains high surfactant and high phosphate content but local soap contains series of potassium, sodium, surfactant and low phosphate content. Seeds of both monocots and dicots observed to have germinated at a particular stage died off, as time progresses. This maybe as a result of toxicity to the seeds. This supports the report of Eze and Ahonsi (1993), that high phosphate concentrations do not support the germination and growth of seedlings. It will interfere with the natural growth process of the seedlings and the seed will not germinate or only very few seeds will germinate because of absent of water. Similar work has been reported on the seeds of the monocot, *Zea mays* by Carl (1998) who stated that detergents which contain high phosphate were toxic to the seedling of *zea mays* and thus have adverse effects on the germination of the seeds.

It interfered with the natural growth process of the seedlings and thus some seeds did not germinate while few of the seeds germinated.

This germination of the seeds observed in these monocots may be as a result of its endosperm or nature of testa.

Contrary to the results obtained with detergents, local soap tends to acts as fertilizer or growth promoter. This is because as time progresses, one hundred percent seed germination was recorded (Figures 1-4). This observation was earlier reported by McBain and Salmon (1970), that a local soap which contains sodium, potassium or low concentration of phosphate acts as a fertilizer for the seeds, and it speeds up the rate at which the seed will germinate unlike that of the high concentration phosphate found in detergents. Waste water from clothes washed with local soap could be used to water crops while waste water from clothes washed with detergent could result to the death of crops.

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EFFECTS OF DETERGENTS AND LOCAL SOAP ON CAJANUS CAJAN

Erhenhi, A.H

*Department of Botany, Delta State University, P.M.B 1, Abraka, Delta State.

E-mail: mac_harrison7@yahoo.com

Abstract

Young plants of pigeon pea, *Cajanus cajan*, were selected and subjected to different concentrations (5%, 10%, 15% and 20%) of detergents (ARIEL, BUNUX and OMO – denoted by the letters **A**, **B** and **O**) and local soap (SODA – denoted by the letter **S**). The results showed that as the concentration of the detergents increased from 15%-20%, growth parameters (plant height, leaf area, girth and leaf number) decreased progressively. Plants treated with local soap “Soda” at different concentrations (5%, 10%, 15% and 20%) had their growth parameters not affected as they grew well like the plants in control (water). Senescence was observed in plants treated with detergents leading to total mortality at 20% concentration. Total mortality was not observed in plants treated with different concentrations of local soap “Soda”.

Key words: Detergents, local soap “Soda”, mortality

INTRODUCTION

Helenius et al., (1979) defines detergent as Substances that when dissolves in water posses the ability to remove dirt form surfaces. Such as the human skin, textiles and other solids, such is termed a detergent.

Detergents can also be defined as amphiphathic molecules that self-associate and bind to hydrophobic surface. Their intrinsic property forming curved micelles in aqueous solution makes them useful for solubilizing planar biological membranes proteins by the formations of mixed micelles often without denaturing them. Although, they have proved invaluable tools for solubilizing integral membrane proteins.

It has become apparent that not all detergent are equally efficient at solubilizing membranes, and that membranes proteins and lipids are differentially extracted by individual detergents Garavito et al., (2001). These observations have provided support to the concept that, membrane are not homogenous and contains micro domains with distinct lipid and protein composition.

The effects of detergent on plants vary depending on how the plant is exposed to it first of all, if a plant is sprayed with detergent solution to cover all the leaves, the detergent which contains surfactant as a component has a lethal effect on the plant Gellini *et al.*, (1985). In general biological detergents are most commonly used to disrupt the bipolar lipid membrane of cells in order to first free on their solubilize membrane-bound proteins. Some detergents can also be used to solubilize recombinant protein, while others find their usage in the stabilization, crystallization, or denaturing of proteins. Additional applications include the extraction of DNA and RNA, the solubilization of specimens for diagnostic application, the lysis of cells, the preparation of liposomes, prevention of reagents and analyze precipitation from solution, and the prevention of non-specific binding in immunoassays Hjelmeland (1990). Detergents have the following components, surfactant, Abrasives, water softener, oxidizers, non-Surfactant, Enzymes and other ingredients among these entire components; surfactant has the most lethal effects on plant (vegetation). Gellini *et al.*, (1985).

The term “soap” refers to particular types of detergent in which the water-solubilized group is carboxylate and the positive ion is usually sodium or potassium. Soap is manufacture by an alkaline hydrolysis reaction called saponification.

MATERIALS AND METHOD

Healthy seeds of pigeon pea, *Cajanus cajan* were obtained from the different markets in Abraka, Delta State. The soil used was sieved with sand sieves (filter) to remove debris and to loosen the soil particles to enable easy percolation of water. The sieved soil was then weighed with weighing balance into nursery bags and each bag weighed 100g. The bags were replicated and three seeds were planted per bag. The plants were watered regularly for germination and stability. The detergents and soap implored are the commonest used domestically (Ariel, Bonux , Omo and Soda) by the people of Nigeria.

Different concentrations were made into 5%, 10%, 5% and 20% of the detergents and soap. Viability test was done to ascertain the viability of seeds for planting into polythene bags.

RESULTS



Plate1 (colour online): Effects of 5% concentration of detergents and local soap on *Cajanus cajan* (pigeon pea) under 2 weeks of treatment

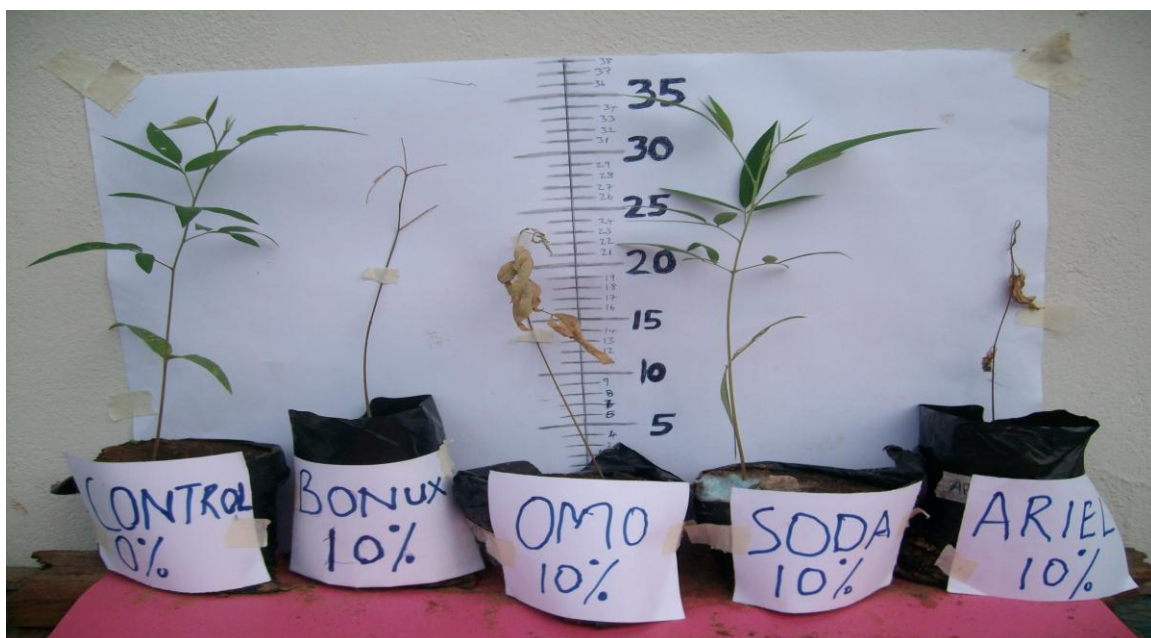


Plate2 (colour online): Effects of 10% concentration of detergents and local soap on *Cajanus cajan* (pigeon pea) under 2 weeks of treatment



Plate3 (colour online): Effects of 15% concentration of detergents and local soap on *Cajanus cajan* (pigeon pea) under 2 weeks of treatment



Plate4 (colour online): Effects of 20% concentration of detergents and local soap on *Cajanus cajan* (pigeon pea) under 2 weeks of treatment

In Plates 1-4, less than a week of treatment, at 5% concentration, plants treated with the three different detergents exhibited senescence which leads to mortality of the plants with exception to Ariel detergent. Plants treated with 5% concentration of local soap did not exhibit senescence and mortality (Plate 1). Total mortality occurred in all the plants treated with 10%-20% concentrations of the three detergents while plants treated with local soap at these concentrations were unaffected (Plates 2, 3 & 4)

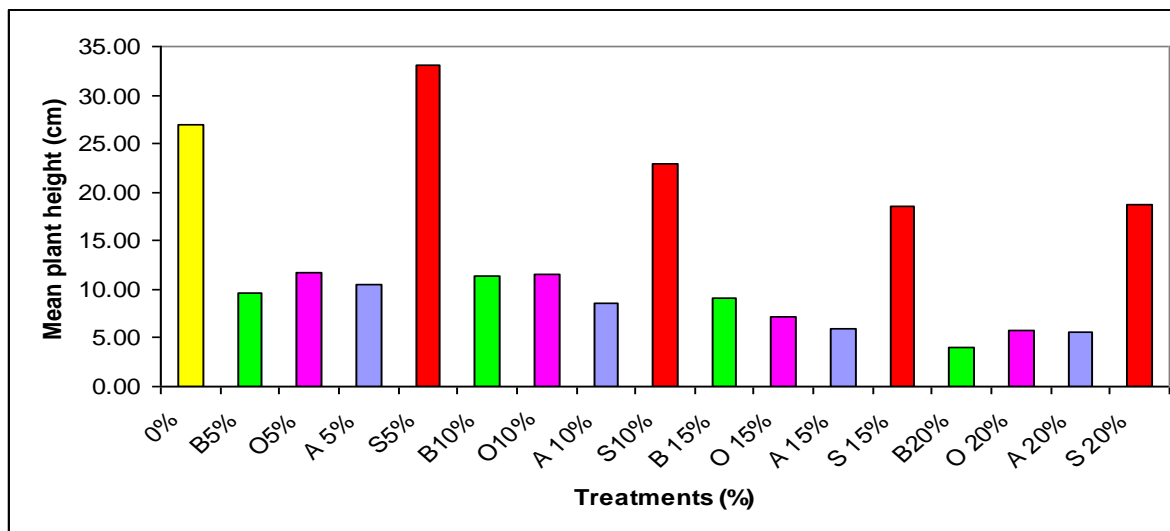


Fig 1 (colour online): Effects detergents and local soap on the Mean plant height of *Cajanus cajan* (pigeon pea) under 2 weeks of treatment.

As the concentrations increased, there was a progressive decrease in the mean plant height. The highest mean plant height was obtained in plants treated with 5% concentration of soda (Fig 1). The mean plant height of plants treated with soda at different concentrations was significantly different ($P < 0.05$) from plants treated with the three different detergents' concentrations (Fig 1)

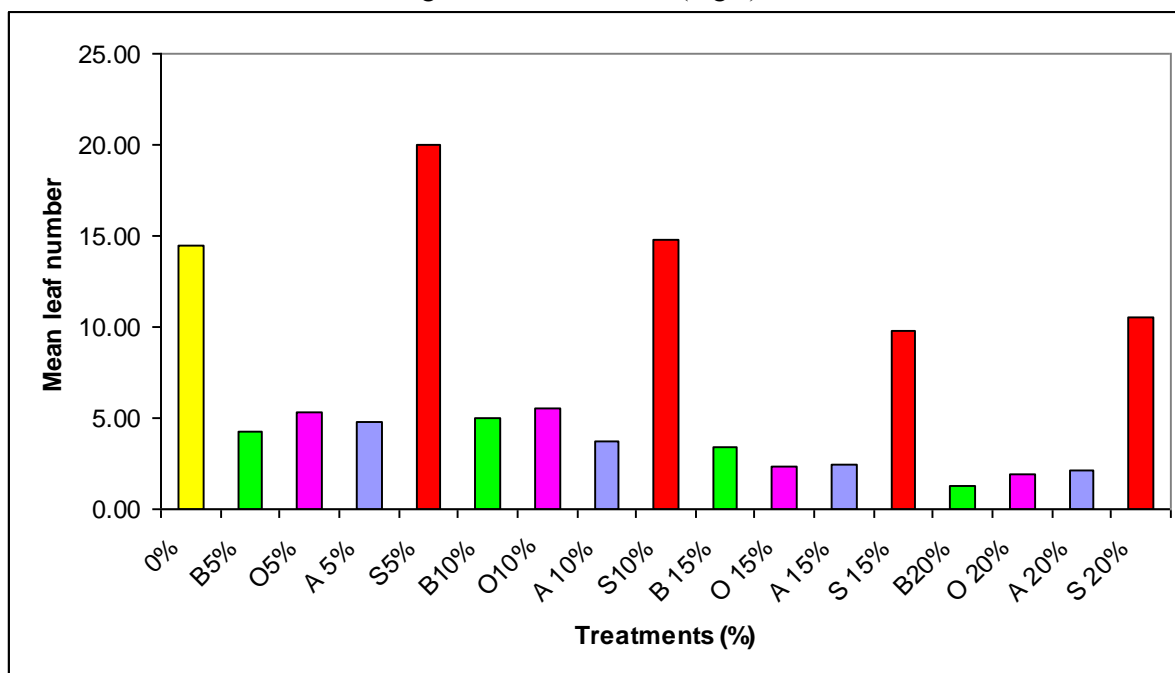


Fig 2 (colour online): Effects detergents and local soap on the mean leaf number of *Cajanus cajan* (pigeon pea) under 2 weeks of treatment.

There was no significant difference ($P > 0.05$) in the mean leaf number of plants treated with 5% concentration of the detergents (Fig 2). Increase in the concentration of both local soap and the detergents resulted in the decrease of the mean leaf number. The highest mean number was obtained in the plants treated with 5% concentration of local soap. It was observed that there was significant difference ($P < 0.05$) between the mean leaf number of plants treated with 5% concentration of local soap and those treated with 20% concentration of the local soap (Fig 2).

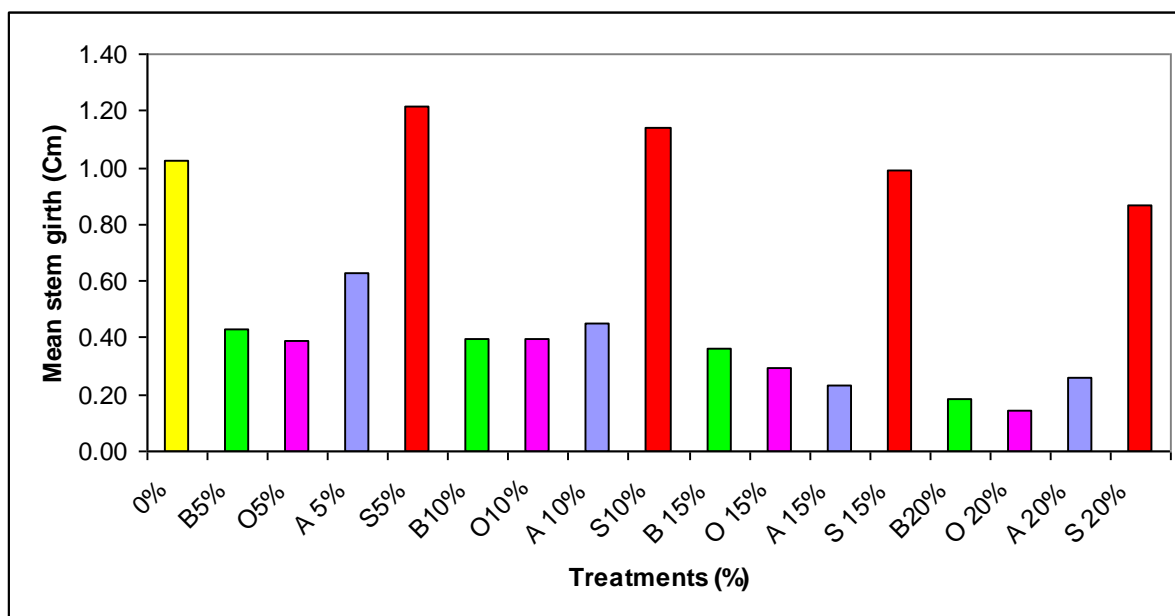


Fig 3 (colour online): Effects detergents and local soap on the mean stem girth(cm) of *Cajanus cajan* (pigeon pea) under 2 weeks of treatment.

As concentrations increased, there was a similar pattern of decrease in the mean girth of all the plants. There was no significant difference ($P > 0.05$) between the mean stem girth obtained in plants treated with 5% and 10% Bonux and Omo detergents. The highest mean stem girth was obtained in plants treated with 5% concentration of local soap. There was no significant difference ($P > 0.05$) in the mean stem girth between plants treated with 5% and 10% concentrations of local soap. The least value of mean stem girth was obtained in plants treated with 20% concentration of Omo detergent, and was found to be significantly different ($P < 0.05$) from the mean stem girth of plants treated with 5% concentration of local soap and control (Fig 3)

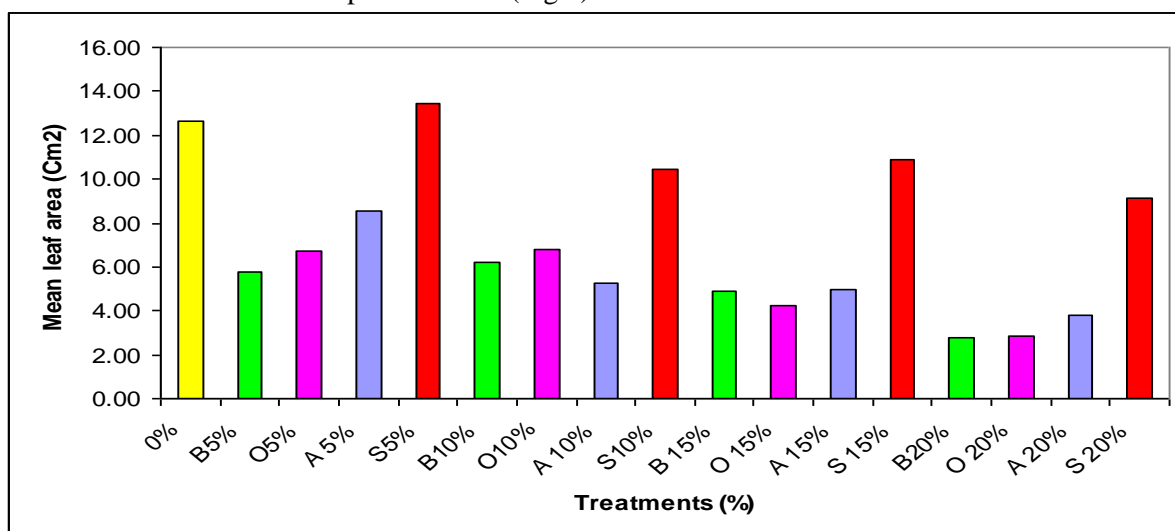


Fig 4 (colour online): Effects detergents and local soap on the mean leaf area (cm²) of *Cajanus cajan* (pigeon pea) under 2 weeks of treatment.

The highest value of mean leaf area was obtained in plants treated with 5% concentration of local soap while the least values of mean leaf area were obtained in plants treated with 20% concentration of Bunux and Omo detergents (Fig 4). There was significant difference ($P < 0.05$) between the mean leaf area of plants treated with 5% concentration of local soap and the plants treated with 20% concentration of Bunux and Omo detergents (Fig 4). There was significant difference ($P < 0.05$) between plants grown in control and those treated with 5%, 10%, 15% and 20% concentrations of the three different detergents (Fig 4).

DISCUSSION

It was observed that low concentration (5%) of local soap “Soda” favoured the growth parameters of plants used (Plates 1-4). This is in accordance with what Carl, (1998) reported that soap at a minute concentration acts as fertilizer to the plant while at higher concentration it then shift from its positive effects to harmful effects to the plant.

As low as 5% concentration of the three different detergents, growth parameters were hindered. The severity of the effects on plants also depends on the level of concentration, although it never had any positive effects on plants. This was earlier reported by Gellini, *et al.*, (1985) that excess detergent to plants has detrimental effects on the vegetation including alternation to stomata and epicuticular waxes.

The inhibitory effects that lead to total mortality in plants treated with detergents started from the least concentration (5%). The higher the concentration, the higher the adverse effects on growth parameters and the effects of detergent on plant are systemic because all parts of the plants are affected (Figs 1-5), this concurs with the previous work by paoletti, *et al.*, (1989) that detergent causes direct injury such as alterations in photosynthesis, shrinking of the stem girth, leaf length, plant height, yellowing of leaves and finally the total mortality of the plants.

This agrees with the earlier report of Gellini *et al.*, (1985) that visible leaves injury has been observed in *Pinus* treated for one week with 100mg detergent. The severity of the detergents on the plants was so high that total mortality was recorded. Large quantities of surfactant which is a component of detergent can cause direct injury on plant such as alteration in photosynthesis inhibition of growth paoletti, *et al.*, (1989) transpiration, Smith (1941), induction of high ratio of chromosome aberration Bellani, *et al.*, (1991) and germ tube elongation, Feder (1981) and also root elongation and mitotic index. Contrary to the results obtained with detergents, local soap tends to act as growth promoter.

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COMPARATIVE EFFICACY OF WATER AND ETHANOL FOR THE EXTRACTION OF PHYTOCHEMICAL CONSTITUENTS OF ALOE VERA SUCCULENT LEAVES

Ilondu, E. M.

Department of Botany, Delta State University, Abraka. Tel. 08036758249. E-mail: martinailondu@yahoo.co.uk

ABSTRACT

The research study was conducted to compare the efficacy of extracting the phytochemical constituents of *Aloe vera* succulent leaves by aqueous method with that of ethanolic method. The analysis was performed using Gas Chromatography-Mass Spectrometry (GC-MS). The result showed that the extracts contained mixture of compounds of which twelve (12) were identified with the amount in ethanol extract being insignificantly ($P=0.05$) higher based on the peak areas.

Keywords: *Aloe vera* extracts, phytochemical constituents.

Abbreviations: GC-MS, Gas chromatography -mass spectrometry; EI, election impacted; NIST, National Institute of Standards and Technology.

INTRODUCTION

Aloe vera (syn. *Aloe barbadensis*) (Plate 1) is a phanerogam (angiosperm) belonging to the family of Liliaceae. It is a perennial drought resistant succulent plant believed to have originated from African continent especially Egypt (Daodu, 2000).

Aloe plant is monocarpic, (flowers once in its life time) and the flowers resemble a small trumpet (Plate 2). The plant contains 96% water and over 75% other constituents which include vitamins, minerals, enzymes, sugars, phenolic compounds, saponins, amino acids etc. (Joshi, 1998). The plant is popularly employed in professional medicine and cosmetics industries (Akindele 2000; Daodu, 2000). Many herbal drugs and drinks have been formulated from *Aloe vera* for maintenance of good health (Davis and Moro, 1989; Loots et al., 2007). It is propagated by suckers (Plate 3) and people now grow *Aloe vera* around their houses.

Aloe vera gel has been reported to be used in ameliorating ultra violet radiation damage (Shelton 1991), sore and wounds, skin cancer, dermatophytic disease, cold and cough, constipation and pile (Regnolds and Dweck 1999; Kafaru, 1994; Hegger, 1996; Daodu, 2000; Djeraba and Quere, 2000; Ilondu and Okoegwale, 2002). The use of Aloe plant in the treatment of other diseases such as asthma, ulcer and diabetes have been reported (Beppu et al., 2003; Tanaka et al., 2006; Rjasekaran et al., 2005; Davis and Moro, 1989). In cosmetic industries, *Aloe vera* is used in the production of toilet soap, hair shampoo, tooth paste and body creams (Daodu, 2000; Akindele 2000)

This study was therefore undertaken to extract and analyze the chemical contents of *Aloe vera* leaves using water and ethanol in order to identify the active chemical constituents therein. It is envisaged that this information would be useful in evaluating the acclaimed wonder works of *Aloe vera* in cosmetics and traditional herbal preparations.

MATERIALS AND METHODS

Collection of plant materials

Fresh healthy *Aloe vera* leaves were collected from *Aloe vera* gardens in Benbo village Ekrejeta, Abraka. The samples were kept in sterile polythene bags and taken to the laboratory for extraction. The leaves were washed three times with sterile distilled water and then macerated.

Extraction method

Aqueous

1 kg of the macerated sample was soaked in 1 litre of sterile distilled water and shaken continuously for 24 h using a rotary shaker (Oyewale and Audu, 2007). The extract was subsequently filtered through four folds of cheese cloth and stored at 4°C until use.

Ethanol

1 kg of the macerated sample was extracted using soxhlet extractor with 95% ethanol. The extract was evaporated on a rotary evaporator at 40°C to remove excess alcohol. Also, the extracts were separately sterilized by passing it through 0.4 µm membrane filter.

Analysis of the extract

The characterization, identification and determination of relative amount of the components of the *Aloe vera* extracts were done using Gas Chromatography – Mass Spectrometry (GC-MS) and GC co-injection of the extracts with authentic standards (Asawalam *et al.* 2008). The analysis was performed on a capillary GC-MS Agilent 6890N equipped with a split capillary injector system DB-5MS, 0.25 mm *30 m* 0.25 µm, maximum temperature of 100°C normal film thickness 0.25 µm, constant flow mode and nominal initial pressure of 3.06 psi.

The carrier gas was helium at a flow rate of 0.5 ml/min. The MS was operated in the Electron Impacted (EI) mode. The preliminary identification of the constituents was based on the computer matching of mass spectral data of the component against the standard NIST (National Institute of Standards and Technology) Library Spectra constituted from spectra of pure substances and components of the known extracts and literature MS data. They were confirmed by their GC retention time comparison with those of reference compounds peak enhancement as well as co-injection/co-elution with authentic standards. The relative proportion of the extract was computed in each case from GC-MS peak areas.

RESULTS AND DISCUSSION

The GC profile of the aqueous and ethanolic extracts are shown in Figures 1 and 2, respectively. The analysis of the extracts revealed a complex mixture of constituents. A total of twelve (12) compounds were identified by comparison of GC-mass spectral fragmentation pattern with those of the NIST

Library (Tables 1 and 2). The extracts represent mainly a mixture of monoterpenes and sesquiterpenes. Their chemical structures are shown in Figure 3.

In aqueous extract, caryophyllene occurred in the largest quantity (502.15 mg/kg). This is followed by linalylpropanoate (192.09 mg/kg) and cyclodecanone (ISTD) (191.17mg/kg). Similarly, cyclodecanone (ISTD) occurred in the largest quantity (357.98 mg/kg) in the ethanolic extract followed by eucalyptol (295.32 mg/kg) and caryophyllene (278.69 mg/kg).

The more the peak area, the more the amount of the component extracted, hence limonene (228.33 mg/kg), eucalyptol (295.32 mg/kg) and cyclodecanone (357.98 mg/kg) were more in the ethanol extracts than in aqueous extract. Only caryophyllene (502.15 mg/kg) and 1-octanol were more abundant in aqueous extract than in the ethanol extracts. α -Pinene, α -phellandrene, borneol, linalylpropanoate, camphene and α -farnesene were of equal amount in both aqueous and ethanol extracts. The abundance of some identified components in the extracts could be attributed to their solubility in the extracting solvent. For example, eucalyptol is insoluble in water but miscible with ethanol (Wikipedia, 2009) hence of higher quantity in ethanol extracts. Borneol, octanol, α -pinene and α -phellandrene are of low solubility hence with lower quantity in the extracts.

Generally ethanolic method of extraction gave the total amount (1680.77 mg/kg) of the compound identified which is not significantly ($P=0.05$) higher than that of the aqueous extract (1526.32 mg/kg). Caryophyllene is a natural bicyclic sesquiterpene used commercially as food additive, in cosmetics and for the treatment of inflammation and pain (Gertsch, et al., 2008) and as a local anaesthesia (Ghelardini et al. 2001). Eucalyptol (1,8 cineole) is used in flavoring fragrances and cosmetics. It is an ingredient in mouth wash and cough suppressant, and controls mucus hypersecretion and asthma via anti-inflammatory cytokine inhibition (Juergens et al., 2003). Eucalyptol has been reported to reduce inflammation and pain when applied topically (Santos and Rao, 2000). Limonene is used in cosmetic products, food manufacturing and medicine. It has been reported to be a significant chemopreventive agent with potential value as a dietary anti-cancer tool in human (Crowel, 1999). Asawalam et al., (2006, 2008) have reported that eucalyptol (1, 8 Cineole), pinene and phellandrene, the constituents of essential oil from *Xylopia actiopica* evoked a high repellent action against *Sitophilus zeamais*. *Aloe vera* phytochemicals have been reported by Rajasekaran et al. (2005) and Loots et al. (2007) for their antioxidant capacity and health benefits. Camphene is used as food additive and flavors while octanol is used in perfumery. α -Farnesene acts as alarm pheromones in termites (Sobotnic et al., 2008).

Conclusion

Aloe vera extracts contained a mixture of constituents and its use in medicine, cosmetics and food industries could be attributed to the properties of these constituents. Both water and ethanol can be used as solvent for extraction of the constituents with little advantage of one over the other.

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Plate 1 (colour online): Healthy *Aloe vera* plant. **Plate 2** (colour online): Flowering plant of *Aloe vera*.



Plate 3 (colour online): *Aloe vera* with suckers.

Table 1. Major identified constituents of *Aloe vera* ethanolic leaf extract and their relative proportions in the extract.

Retention time (min)	Height (pA)	Amount (mg/kg)	Name of compound	Molecular formula
6.36	10.08	31.35	α -pinene	C ₁₀ H ₁₆
7.28	6.95	21.63	β -myrcene	C ₁₀ H ₁₆
7.93	73.37	228.33	Limonene	C ₁₀ H ₁₆
8.04	94.90	295.32	Eucalptol	C ₁₀ H ₁₈ O
8.40	17.04	53.02	α -phellandrene	C ₁₀ H ₁₆
8.88	5.58	17.36	1-octanol	C ₈ H ₁₈ O
10.11	3.28	10.10	Borneol	C ₁₀ H ₁₈ O
10.47	61.73	192.09	Linalylpropanoate	C ₁₃ H ₂₂ O ₂
12.32	115.03	357.98	Cyclodecanon (ISTD)	C ₁₀ H ₁₈ O
13.71	89.56	278.69	Caryophyllene	C ₁₅ H ₂₄
13.85	34.72	108.03	Camphene	C ₁₀ H ₁₆
14.75	27.89	86.77	α -Farnesene	C ₁₅ H ₂₄
				1680.77

Table 2. GC-MS phytochemical compounds identified from *Aloe vera* aqueous leaf extracts.

Retention time (min)	Height (pA)	Amount (mg/kg)	Name of compound	Molecular formula
6.36	10.08	31.35	α -pinene	C ₁₀ H ₁₆
7.28	6.95	21.63	β -myrcene	C ₁₀ H ₁₆
7.93	48.34	150.42	Limonene	C ₁₀ H ₁₆
8.04	48.11	149.72	Eucalptol	C ₁₀ H ₁₈ O
8.40	17.04	53.02	α -Phellandrene	C ₁₀ H ₁₆
8.88	9.57	29.77	1-octanol	C ₈ H ₁₈ O
10.11	3.28	10.10	Borneol	C ₁₀ H ₁₈ O
10.47	61.73	192.09	Linalylpropanoate	C ₁₃ H ₂₂ O ₂
12.32	61.44	191.17	Cyclodecanon (ISTD)	C ₁₀ H ₁₈ O
13.71	161.37	502.15	Caryophyllene	C ₁₅ H ₂₄
13.85	34.72	108.03	Camphene	C ₁₀ H ₁₆
14.75	27.89	86.77	α -Farnesene	C ₁₅ H ₂₄
				1526.32

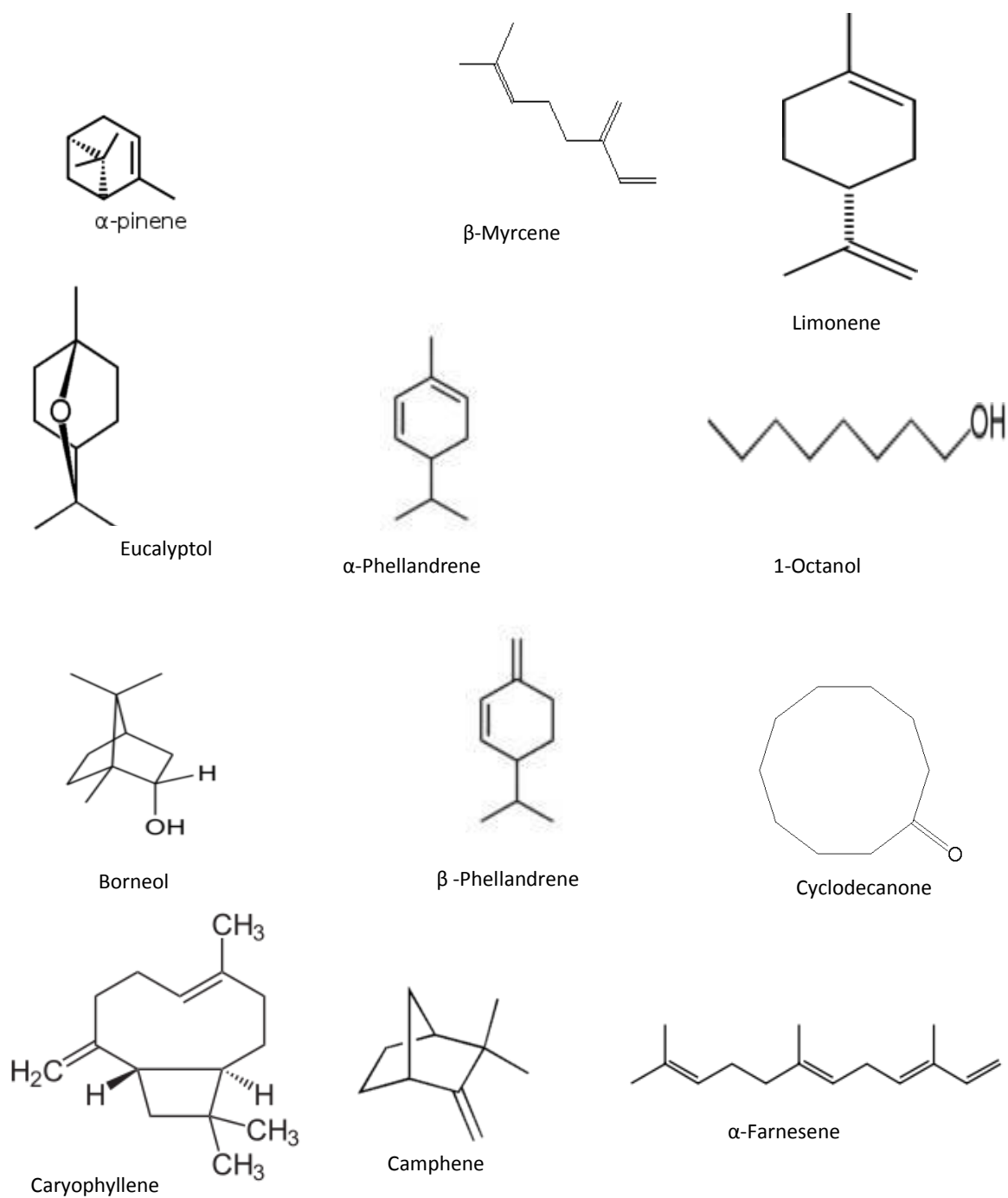


Figure 3. Chemical structures of identified constituents of Aloe vera extracts.

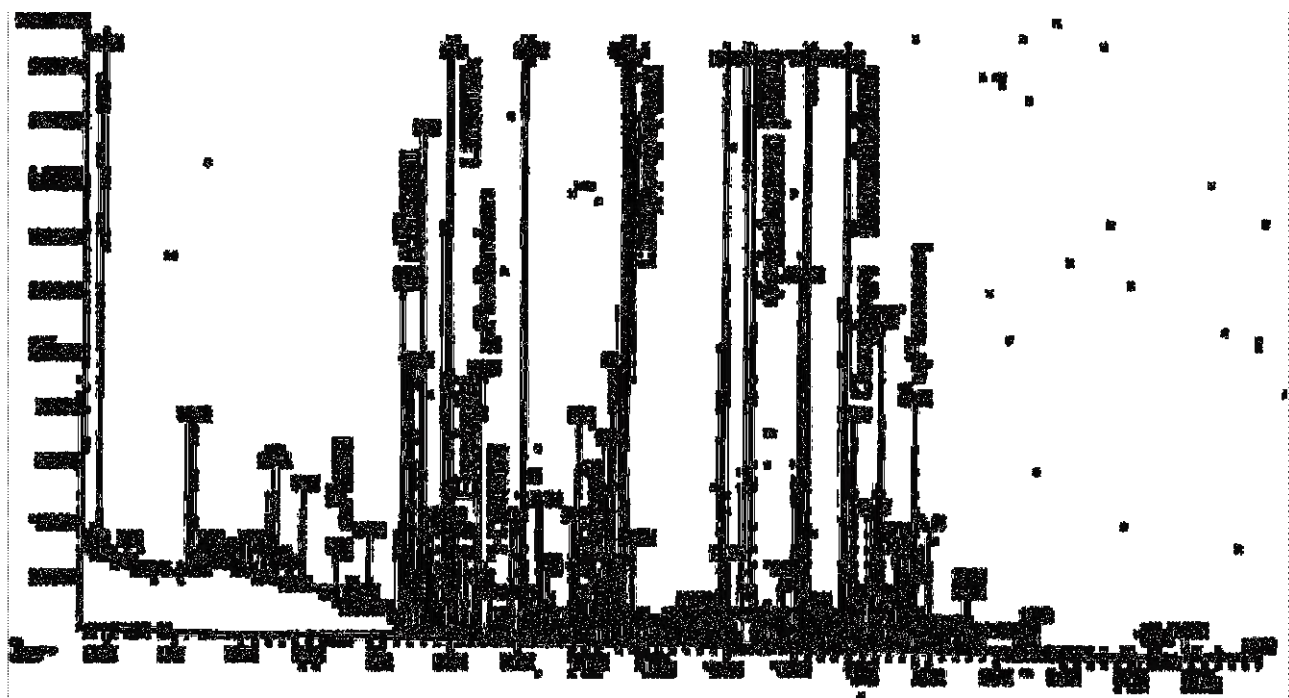


Figure 1. GC-MS chromatogram of *Aloe vera* ethanolic leaf extract.

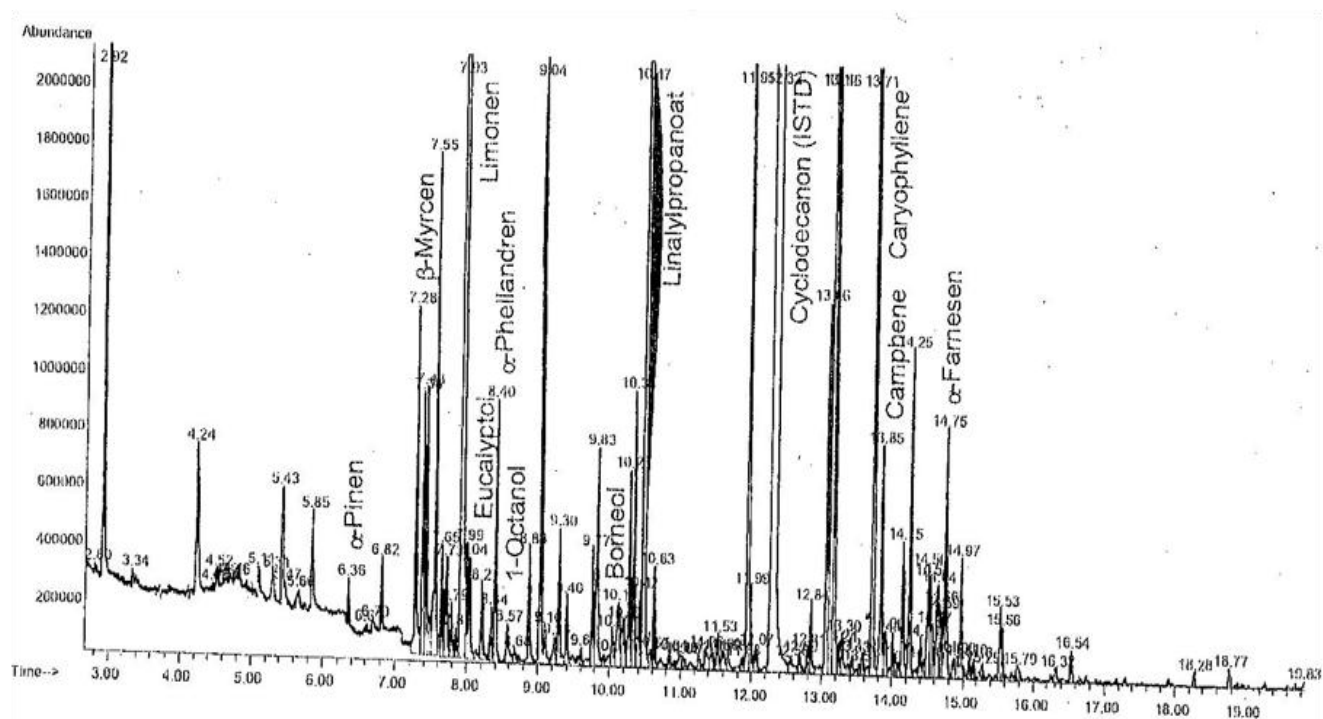


Figure 2. GC-MS chromatogram of *Aloe vera* aqueous extract.

**IN-VITRO EVALUATION OF FOUR FUNGICIDES FOR THE CONTROL OF
SCLEROTIUM ROLFSII SACC., THE CAUSAL AGENT OF RHIZOME ROT OF GINGER
(ZINGIBER OFFICINALE ROSC)**

Ilondu, E. M.

Department of Botany, Delta state University Abraka.
. E-mail: martinailondu@yahoo.co.uk.; Phone. 08036758249

Abstract

The efficacy of four fungicides (Benlate, Dithane M₄₅, Funguran-OH and Team) for the control of the radial growth, sclerotia production and germination of *Sclerotium rolfii* isolated from rotted ginger rhizome was evaluated *in-vitro*. Result revealed that all the fungicides significantly ($P < 0.05$) inhibited the activities of *S. rolfii* in varying degrees. The effectiveness of the fungicides increased with an increase in concentration. Team and Dithane M₄₅ completely inhibited the growth and sclerotia production of the test fungus at 200 and 300 ppm, respectively, while maximum inhibition was achieved by Benlate and Funguran-OH at 1000 ppm. The fungicides inhibited germination of sclerotia in the following order: Team>Funguran-OH>Dithane M₄₅>Benlate

Abbreviations: WA, Water agar; PDA, potatoe dextrose agar.

Keyword: *Sclerotium rolfii*, growth inhibition, fungicides.

INTRODUCTION

Ginger is an underground stem (rhizome) of *Zingiber officinale* Rosc (Plate I), a herbaceous perennial in the family of Zingiberaceae. About 700 species are recognized but two varieties (Yellow and Black ginger) are grown in Nigeria (Arene et al., 1986). As a spice, ginger is an aromatic vegetable product of tropical origin used in a pulverized state for seasoning or garnishing food and beverages (Sammbamurty and Subrahmanyam, 1998; Afshari et al., 2007). Ginger prevents flatulence and aids digestion. It is also a stimulant, carminative, diaphoretic, pungent, sialogogue and condiment (Kafaru, 1994; Ernst and Pitler, 2000; Jaw-Chyun, et al., 2007). Rhizome rot (plate 2) also known as soft rot of ginger is quite severe in ginger growing areas including Nigeria and is caused by a number of organisms including *Sclerotium rolfii* (Mehrotra and Aggarwal 2003).

S. rolfii is a ubiquitous soil inhabiting fungus (Omenka, 2001) with extensive host range (Maduewesi, 1975; Wokocha, 2001) and attacking many crops including yams, tomatoes, cassava, cowpea, pepper, okra, etc. causing rots (Ejechi and Ilondu, 1999; Wokocha and Okereke, 2005; Wokocha, 1998) and damping off of seedlings (Arinze and Maduewesi 1989; Onuegbu and Brown 1992). Sclerotia are the over-wintering structures and therefore serve as the major source of infection. The ability of *S. rolfii* to rapidly attack and colonize a large number of plant species has been attributed to the production of both macerating enzymes and oxalic acid (Mehtotra and Aggarwal, 2003). There is no single control measure for *S. rolfii*. However, many agrochemicals have been recommended for the control of this fungus.

This work is therefore undertaken to evaluate the efficacy of four fungicides in controlling the growth and germination of *S. rolfii* *in vitro*. It is hoped that identifying and applying an effective fungicide would help to limit the viability of *S. rolfii* in the environment.

MATERIALS AND METHODS

Isolation and preparation of pure culture

Partially rotted rhizome of *Z. officinale* (plate 2) was used in isolating *S. rolfsii*. The fungus appeared as a mat of whitish mycelium on the surface of the wounded rhizome. Sections (2-4mm long) excised from the rotted rhizome with sterile razor blade were surface-sterilized for two minutes in 2% aqueous solution of commercial bleach and rinsed in two changes of sterile distilled water. The disinfected tissue pieces were blotted between sterile Whatman No. 1 filter papers and aseptically plated in 9 cm diameter Petri-dish containing water agar (WA). The setup was incubated at room temperature ($30\pm 2^{\circ}\text{C}$) on laboratory bench for 3-5 d (Wokocha and Okereke, 2005). The mycelium growing from the diseased tissue on WA was transferred to potato dextrose agar (PDA) plates amended with 1.0 mg/ml chloranphenicol to check bacterial contamination (modified PDA). The plates were incubated for 5-7 d and sub-cultured repeatedly unto clean PDA plates until pure cultures were obtained (Plate 3). Pure cultures were also obtained through the use of small sclerotia already developed on the surface of the rotted rhizome (Maduwesi, 1975). The cultures were stored for sclerotia production for subsequent studies.

Preparation of the fungicide-potato dextrose agar

Four fungicides (Table 1) were evaluated for their efficacy against *S. rolfsii*. These fungicides were selected because they are readily available and were procured from Delta State Agricultural procurement Agency (DAPA) Ibusa, near Asaba. Measured quantities of the fungicides were added into 500 ml conical flasks to prepare the stock solution for each fungicide and for each level of concentration. The levels of concentration for each fungicide used were 5, 10, 25, 50, 75, 100, 200, 300, 500, 750 and 1000 ppm of the active ingredient (Odigie, 2000). One milliliters of each level of concentration was aseptically incorporated into 20 ml of cool molten modified PDA in each of the test tubes. Each medium was thoroughly homogenized by gentle agitation for uniform dispersal of the fungicide before dispensing into 9 cm diameter sterile Petri-dishes. The modified PDA without the fungicide represents the control. The plates were allowed to set on the laboratory bench for 6 h.

Growth and germination of *S. rolfsii*

With the aid of a sterile cork borer, each Petri dish was inoculated by placing 4 mm mycelial disc taken from the periphery of actively growing 4 d old culture of *S. rolfsii*, face-downward at the centre of the Petri dish. The centre was determined by drawing two perpendicular lines on the bottom of the Petri-dish before dispensing the medium. Three replicate plates of each concentration of each fungicide and their control were incubated at room temperature ($30\pm 2^{\circ}\text{C}$) on the laboratory bench for 7 d in a completely randomized design (Nwanosike and Adeoti, 2002). The fungi toxicity was evaluated after 7 d by measuring the colony diameter, taking the average of the horizontal and vertical growth of the mycelia with a transparent metric ruler. The experiment was repeated twice and was terminated each time when the radial growth of the fungus reached the rim of the Petri dish in the control plates. The percentage inhibition of the mycelial growth was calculated following Wokocha and Okereke (2005).

$$FP = \frac{dc-dt}{dc} \times \frac{100}{1}$$

where, FP, Percentage inhibition of fungal growth; dc, average diameter of fungal colony in control Petri dish; dt, average diameter of fungal colony in treated Petri dish.

The sclerotia produced in each level of concentration of each fungicides were counted two weeks after inoculation and recorded. On sclerotia germination studies, 30 brown sclerotia of uniform size and age were selected for each fungicide concentration. The sclerotia were dipped into sterile distilled

water and later inoculated into different concentrations of the fungicides in the Petri dish and replicated 3 times as described above. They were observed for germination after 3 d (Onuegbu et al., 2001). The number of germinated sclerotia was recorded and the percentage inhibition of germination was computed as stated above.

Data analysis

The results were subjected to statistical analysis using two-way analysis of variance (ANOVA) to test for level of significance between the various fungicides and concentrations. All analysis were performed using the software programme Graph Pad Prism® Software (version 5.0, San Diego, CA).

RESULTS AND DISCUSSION

The result of the *in vitro* evaluation of four fungicides against radial mycelia growth of *S. rofsii* is presented in Table 2. All the fungicides significantly ($P < 0.05$) inhibited the activities of *S. rofsii* in varying degrees. The effectiveness of the fungicides increased with an increase in concentration of each. Team and Dithane M₄₅ were the most effective in reducing the mycelial growth of the fungus, causing a 100% reduction at 200 and 300 ppm respectively. Benlate when applied at 1000 ppm gave the maximum reduction of 76% as against that of funguran-OH which gave 45% (Figure 1). In general, Team, Dithane M₄₅ and Benlate were effective in reducing the mycelia growth while funguran-oH was less effective.

The result of the performance of the fungicides on sclerotia germination of *S. rofsii* is presented in Table 3. None of the fungicides completely inhibited the sclerotia germination on the agar plates even at 1000 ppm concentration. Team gave 93% reduction, funguran-OH gave 90% while Dithane M₄₅ gave 80% reduction with no significant difference ($p < 0.05$) in their over all performance (Figure 2), but there is significant difference ($P > 0.05$) between the performance of Benlate and the other three fungicides. Reduction in radial mycelial growth or total inhibition of the mycelia affected the sclerotia production adversely. There is significant difference ($P > 0.05$) among the sclerotia production in four fungicides tested (Table 4).

Team and Dithane M₄₅ were the most effective in reducing the growth and sclerotia production of *S. rofsii* on PDA amended with fungicides. Jayansinghe and Wijesyndera (1995) reported that 100% inhibition of mycelial growth is considered effective dosage of fungicides. Benlate was also found to be nearly as effective as team and Dithane M₄₅. Team, Funguran-OH and Dithane M₄₅ were also found to be very effective in inhibiting the sclerotia germination. Dithane M₄₅ (Zinc manganese ethylene bisdithiocarbamate) is a broad spectrum fungicide recommended by the manufactures to be effective against many fungi that attacks crops. Odigie (2000), Adeoti (2000) and Nwanosike and Adeoti (2002), have reported that Dithane M₄₅ was very effective in inhibiting the mycelia growth of *Curvularia clavata*, *Coniella musacansis* and *Alternaria macrospore* respectively. Also, Oke (1990) had reported that Dithane M₄₅ was very effective against *Corynespora cecisilola* on tobacco.

The effectiveness of the fungicides could be due to the unique combination of zinc managanese ethylene bisdithiocarbanate ion which provides a more potent fungicide. It could also be due to direct close contact between the fungus and the fungicide in the agar plates (Adeoti, 2000). Team is a dual action (contact and systemic) fungicide and also a combination of mancozeb and carbandazim. This could have been responsible for its effectiveness. Benlate was not as effective as Team and Dithane M₄₅ because it is relatively insoluble in water (Nnodu and Okwuowulu, 1990). Funguran-OH was not very effective in inhibiting the radial mycelia growth probably because appropriate concentrations were not used.

Conclusion

Team, Dithane and Benlate were found to be effective in the control of *S. roffsii* *in vitro*. In lieu of an alternative effective control of *S. roffsii*, Team is highly recommended as it is readily available, a dual action fungicide (topical and systemic) in nature and very economical for use since at lower concentrations, total control of the pathogen was achieved. These fungicides can serve as agents for inoculum reduction in the field if applied as soil drench.

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Table 1. Common Name, Trade Name, Chemical Name, and Formulation of the fungicides evaluated *in vitro* for their efficacy against *Sclerotium. rolfsii*.

Common name of fungicide (ai)	Trade name	Chemical name	Formulation
Benomyl	Benlate	Methy-1-butylcarbomyl-2-benzimidazole-2-yl carbarmate	50% WP
Carbendazim + mancozeb	Team	Mythlybenzimidazole-2yl carbarmate + zinc manganese ethylene bis dithiocarbamate	12 + 63% WP
Mancozeb	Dithane M ₄₅	Zinc manganese ethylene bis dithiocarbamate	75% WP
Cupric hydroxide	Kocide	Funguran-OH	50% WP

WP, Wettable powder; ai, active ingredient.

Table 2. Mean radial mycelial growth (cm) of *Sclerotium rolfsii* at various concentrations of fungicides after seven days of incubation on agar plates.

Conc. (ppm)	Fungicide			
	Benlate	Team	Dithane M ₄₅	Funguran-OH
0	4.00±0.03	4.00±0.00	4.00±0.00	4.00±0.00
5	4.00±0.03	3.93±0.07	4.00±0.00	4.00±0.00
10	3.73±1.18	3.87±0.13	4.00±0.00	4.00±0.00
25	3.33±0.24	3.73±0.07	4.00±0.00	4.00±0.00
50	3.00±0.46	2.93±0.07	3.27±0.18	3.93±0.07
75	2.77±0.38	1.87±0.73	2.70±0.32	3.63±0.22
100	2.37±0.37	0.13±0.23	0.90±0.49	3.27±0.27
200	1.93±0.07	0.00±0.00	0.20±0.10	3.17±0.32
300	1.70±0.10	0.00±0.00	0.00±0.00	3.03±0.29
500	1.53±0.07	0.00±0.00	0.00±0.00	2.90±0.30
750	1.30±0.15	0.00±0.00	0.00±0.00	2.67±0.12
1000	1.00±0.03	0.00±0.00	0.00±0.00	2.20±0.12

Table 3. Mean number of sclerotia germination of *Sclerotium rolfsii* at various concentrations of fungicides after seven days of incubation on agar plates.

Conc. (ppm)	Fungicide			
	Benlate	Team	Dithane M ₄₅	Funguran-OH
0	10.00±0.00	9.33±0.67	10.00±0.33	10.00±0.00
5	10.00±0.00	8.00±1.00	8.33±0.00	9.00±0.58
10	10.00±0.00	6.67±0.33	7.67±0.33	8.67±0.33
25	9.67±0.33	6.67±0.33	7.00±0.33	8.67±0.33
50	9.67±0.33	6.00±0.58	6.67±0.33	6.67±0.88
75	9.67±0.33	5.67±0.33	5.67±0.33	5.33±0.33
100	8.33±0.33	5.00±0.00	5.33±0.33	5.00±0.00
200	8.00±0.58	5.00±0.00	5.33±0.33	4.00±0.58
300	7.00±0.58	3.67±0.67	4.67±0.33	3.67±0.33
500	6.00±0.00	3.00±1.00	3.67±0.33	2.33±0.33
750	5.67±0.33	2.67±0.67	2.67±0.33	1.33±0.33
1000	4.33±0.33	0.67±0.33	2.00±0.00	1.00±0.00

Table 4. Mean number of sclerotia production by *Sclerotium rolfsii* at various concentrations of Fungicides after seven days of incubation on agar plates.

Conc. (ppm)	Fungicide			
	Benlate	Team	Dithane M ₄₅	Funguran-OH
0	97.33±25.44	80.33±10.40	51.67±12.14	48.67±6.67
5	34.67±5.46	24.33±3.93	33.00±8.51	20.67±0.33
10	30.00±3.61	18.67±1.76	22.33±5.49	19.00±1.00
25	25.33±3.33	15.33±1.45	15.33±1.86	17.33±2.67
50	18.33±4.84	12.33±1.33	13.00±1.56	15.67±2.33
75	15.67±2.73	6.00±1.33	12.33±1.45	13.33±1.67
100	11.33±0.67	1.67±1.67	11.33±1.76	11.33±0.67
200	10.00±0.00	0.00±0.00	6.00±1.53	8.33±0.33
300	9.33±0.67	0.00±0.00	0.00±0.00	6.67±0.67
500	9.00±0.58	0.00±0.00	0.00±0.00	5.00±0.00
750	8.00±0.00	0.00±0.00	0.00±0.00	5.00±0.00
1000	5.00±0.58	0.00±0.00	0.00±0.00	4.00±0.00

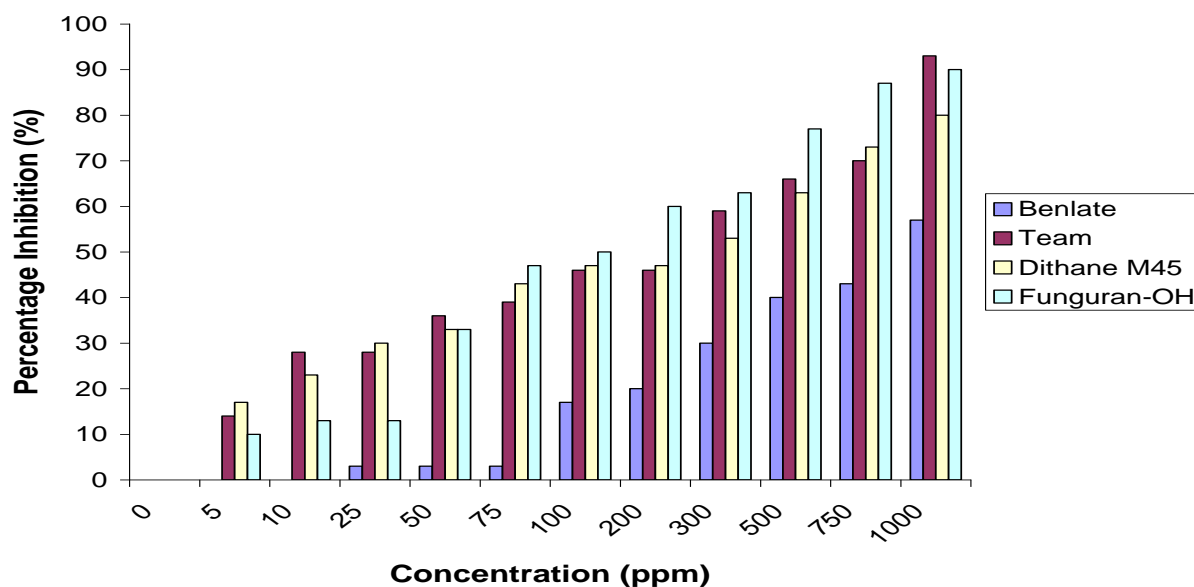


Figure 1 (colour online): Mean percentage inhibition of radial mycelial growth of *Sclerotium rolfsii* at various concentrations of fungicides after seven days of incubation on agar plates.

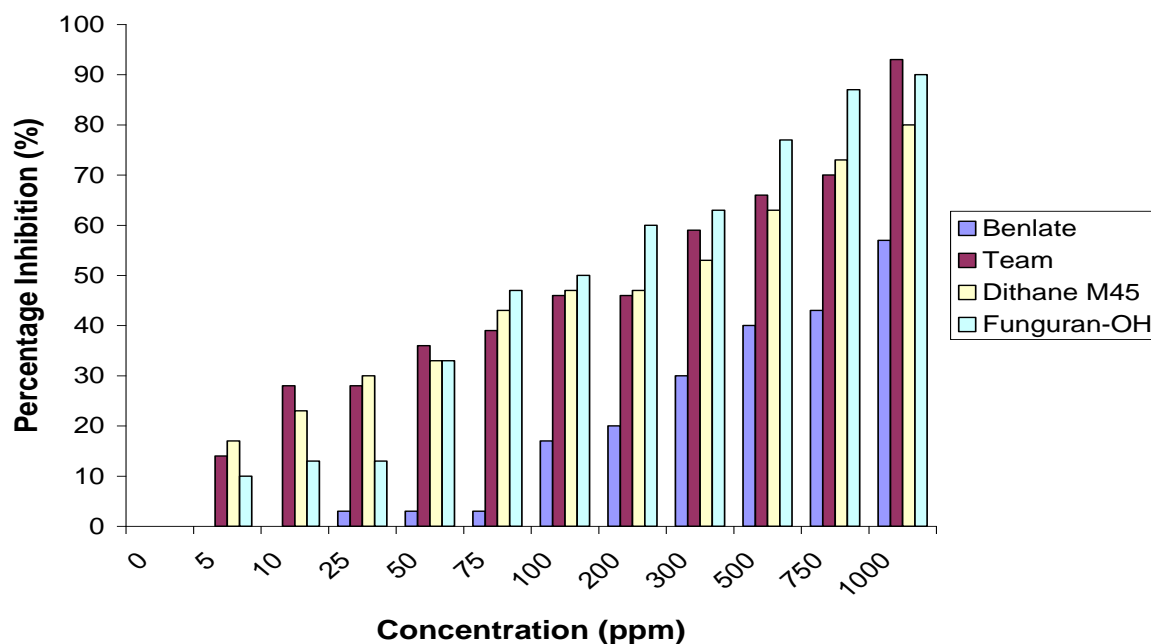


Figure 2 (colour online): Percentage Inhibition of Sclerotia Germination of *Sclerotium rolfsii* at various concentrations of fungicides after seven days of incubation on agar plates.

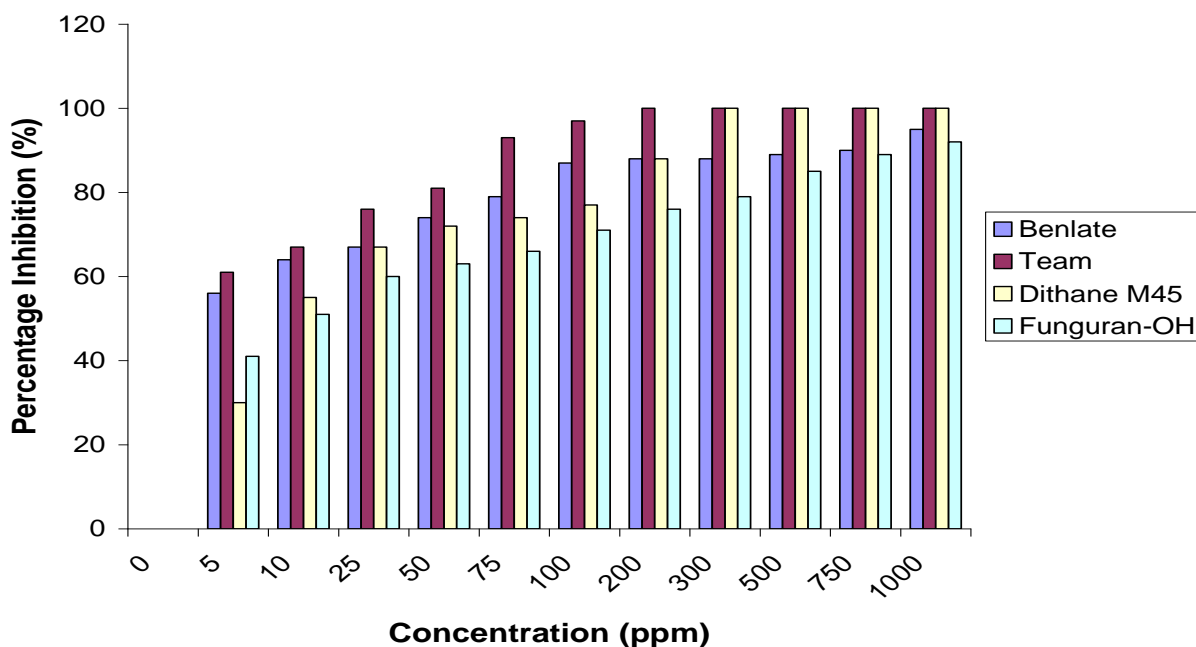


Figure 3 (colour online): Mean Percentage Inhibition of Sclerotia Production of *Sclerotium rolfsii* at various concentrations of Fungicides after seven days of incubation on agar plates.



Plate 1 (colour online): Healthy ginger rhizome.



Plate 2 (colour online): Ginger rhizome affected by *Sclerotium rolfsii* rot.

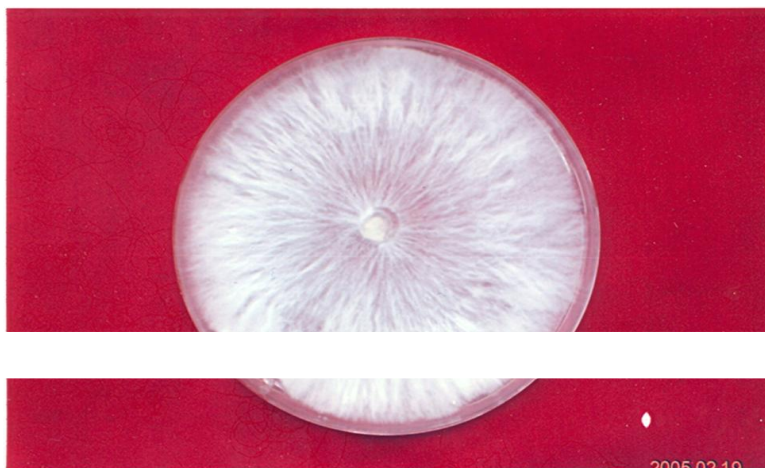


Plate 3 (colour online): Culture of *Sclerotium rolfsii* in a Petri dish.

INTESTINAL HELMINTHS INFECTION IN SCHOOL CHILDREN AND SANITARY CONDITIONS OF PRIMARY SCHOOLS IN AGBOR, DELTA STATE NIGERIA

¹Oduma E. .O. and ²Ogbe M.G

¹DEPARTMENT OF BIOLOGY, COLLEGE OF EDUCATION, AGBOR.

²DEPARTMENT OF ANIMAL AND ENVIRONMENTAL BIOLOGY, DELTA STATE UNIVERSITY, ABRAKA

¹Email: lobito8z@yahoo.co.uk: Phone No: 08037331303

ABSTRACT

A study on intestinal helminthes infection and sanitary conditions was conducted in four primary schools in Agbor metropolis in Ika South Local Government Area of Delta State, Nigeria. A total of 322 samples comprising of 163 males and 159 females aged between 5-13 years were examined using the direct smear method and kato-katz techniques. A child's form and school based questionnaire were used to obtain personal data of pupils and sanitation situation in the schools. The intestinal helminthes detected were *Ascaris lumbricoides* 41 (12.73%), hookworm 28 (8.7%) and *Trichuris trichiura* 16 (5.00%). Age prevalence revealed the 8-10 years age group had the highest prevalence of (33.3%). Sex prevalence showed that males (34.4%) were more infected than their female (18.7%) counterparts ($p < 0.05$). Prevalence and intensity patterns showed significant variations among pupils who had access to better sanitation facilities ($P < 0.05$). Provision of clean and constant water supply, good toilet facilities, health education intervention and adequate sanitation could reverse the trend observed in this study.

INTRODUCTION

Helminthes are known to cause a lot of morbidity and socio-economic deprivation in population, where poor sanitary condition provide optimal environment for their development and transmission. Intestinal helminthiasis is among the most common communicable disease of school-age children in developing countries of the tropics. It also tends to occur at highest intensity in this age group (Albonico *et al.*, 1993).

The health of school age children in developing countries is a concern that has received increasing attention in recent past, following high morbidity rates due to parasitic diseases, which are preventable. Although, death rates among these children are low, it has been estimated that school age children experience a considerable worm burden, which may have both immediate and long term consequences for their healthy growth and education (Bundy and Guyatt, 1996). Often reported cases of intestinal helminthes are *Ascaris lumbricoides*, *Trichuris trichiura* and hookworm (Montessor *et al.*, 1998).

Transmission of these parasites is sometimes influenced by differences in environment, local population and socio-cultural habits such that parasite distribution differ markedly especially among school aged population (Akogun and Badaki, 1998).

Sanitation, a condition that affects human health especially with regard to dirt, infectious agents, specifically disposal of sewage and other domestic wastes from houses tends to lag behind in the developing world. Adequate clean water supply, and safe disposal of excreta is a fundamental importance not only for health of a community but also because of the social and environmental benefits it brings. Poor domestic and personal hygiene involving food and hands often diminishes or even neglect any positive impact of improved excreta disposer on the community. It increases

helminthes infection because these infections are mostly excreta related being dependent on faecal oral-transmission.

Montessoro *et al.*, (1998) recommended that any programme aimed at controlling morbidity due to intestinal helminthes should begin with a baseline survey. Baseline data provides a sound basis for estimating current status and needs for future interventions. It also provides data to guide the developments of control programmes at district, regional and national levels.

This study was therefore undertaken to assess the prevalence and intensity of intestinal helminthes infection among school children in Agbor metropolis. It is also aimed at assessing the sanitary conditions of primary schools in the metropolis.

MATERIALS AND METHOD

The Study Area: Agbor metropolis is made up of Boji Boji Owa and Boji Boji Agbor in Ika North and Ika South Local government area of Delta State. It is located within latitude 6°10' - 6°18' and longitude 6°10'0" - 6°15'0" E of the Greenwich meridian. It is found within the tropical rainforest belt in Nigeria. The area has a relatively high temperature ranging from 25°C to 27°C in the wet season but rises a little to between 27°C to 30°C during the dry season. The metropolis is characterized by an undulating landscape with pockets of hills and slope. It experiences heavy flooding during the rainy season resulting in gully erosion. The major drainage system is the Orogodo River. The inhabitants of this metropolis are civil servants, traders, farmers, artisan workers and transport workers. Sources of water supply in the community are; collected concrete tanks constructed in the ground, pipe borne (tap water), which runs occasionally and borehole water. Latrine facilities include; water closets, pit latrines, while others defecated in nearby bush, sometimes in well dug out open trenches. The heavy surface runoff occasionally increases during the rainy season increases sanitation problems.

Collection of Specimen: Four primary schools namely, Orogodo primary school, College of Education Staff Model primary school, Hedson primary school and Owanta primary school were selected for the study. The study was carried out between December 2004 and May 2005.

Dates for collection of stool samples were arranged with the school heads. Letters of introduction were given to the children for their parents to inform them about the nature of study and their consent. Marked stool containers were then distributed to the children. The containers were retrieved from the pupils the following day. A total of three hundred and twenty-two stool samples were collected from the four schools. A school based questionnaire and a child's form were used to determine the health and sanitation situation in the schools as well obtain information from the individual child.

Parasitological Examination: The stool samples were examined microscopically for consistency, mucous, blood and visible parts of helminthes using methods described by (WHO, 1994). The samples were examined within 4-12 hours after collection. The direct smear method was used to check for the presence of eggs of helminthes microscopically at x10 and x40 magnification, whereas the kato-katz technique was used to measure the intensity of infection (WHO, 1994). A small mould of faecal matter is sieved through a nylon screen. The sieved material was placed on a slide in a template (contains 41.7mg of faeces) using a flat spatula. The template is carefully removed from the slide. The faecal matter on the slide is covered with cellophane strips pre-soaked in glycerol. The prepared slides were kept for one or more hours at room temperature to clear the faecal matter. The number of eggs of each species was then multiplied by 24 to obtain the number of eggs per gram (epg). The egg per gram gives an estimate of worm burden and allows for identification of individuals likely to suffer from severe consequences of infection. Any sample egg contributes to the estimate of prevalence.

The following thresholds for classification of individuals were used for intensity of infection in the study.

	Light intensity infection	Moderate intensity infections	Heavy intensity infections
<i>A.lumbricoides</i>	1-4,999epg	5,000-49,999epg	>50,000epg
<i>T.trichiura</i>	1-999epg	1000-9,999 epg	>10,000 epg
Hookworm	1-1,999 epg	2,000-3,999 epg	>4,000 epg

WHO, (1987)

Data Analysis: The data analysis was done by using the chi-square test.

RESULTS

Table 1, shows the prevalence of intestinal helminthes among the four primary schools in Agbor metropolis. Of the 322 school pupils examined 85(26.4%) were infected with intestinal helminthes. The prevalence rate for the various intestinal helminthes observed was generally low as 12.7% were infected with *A. lumbricoides* 8.7% infected with hookworm and 5% infected with *Trichuris trichiura*. Though intestinal helminthes infection was higher in Orogodo Primary school which recorded a prevalence rate of 31.1% as against the 22.4% in staff model, 28.6% in Owanta and 22.2% in Hedson primary schools there was however no significant difference in infection rate between the schools. Hedson and staff model primary schools had better access to sanitation facilities (Table 6).

Results in Table 2 showed that males (34.4%) were more infected than their female (18.2%) counterparts. The difference in prevalence rate was statistically significant ($P<0.05$). Hookworm prevalence was significantly higher ($P<0.05$) in males than the females.

Results in Table 3 showed the highest intestinal helminthes infection rate was observed among the 8-10years (33.3%) age group, while the least prevalence was among the 11-13years (18.4%). This was however not statistically significant ($P>0.05$).

The prevalence of intestinal helminthes infection varied according to access to better facilities such as water supply and excreta disposal (table 4 and 5). Prevalence rate was least among children whose families obtain water from boreholes (14.9%) than those who obtained water from rainwater stored in concrete tanks (31%). Those who used water closet had a prevalence rate of (19.7%) while the pit latrine users had a prevalence rate of 32%. These differences were statistically significant for hookworm infections ($p<0.05$).

The intensity of infection was light for all the intestinal helminthes observed (fig1).

DISCUSSION

The study showed an overall prevalence of 26.4% (Table 1) among primary school children in Agbor metropolis. Conversely, the present prevalence rate is low when compared with earlier studies among school children in Rumde, Yola, Adamawa State by Gindiri and Akogun, (2000) and among children in areas of operation by shell petroleum of Nigeria Western Division by Ogbe *et al.*, 2002. However a lower prevalence of intestinal helminthes infection was observed in parts of Niger Delta by Okafor and Azuibike, (1992) and Agi, (1995). These prevalence rates have been attributed by many authors to improper hygiene, poor sanitary conditions, agricultural habits and poverty (Scolari *et al.*, 2000, Morales *et al.*, 2003).

Of all the intestinal helminthes observed, *Ascaris lumbricoides* was the most common with a prevalence rate of 12.73%. Similar observations were made earlier by Adeyeba and Akinlabi, (2002) and Ogbe *et al.*, (2002)) with a considerable morbidity among school children.

The low infection rate of hookworm in this study could be attributed to climatic conditions at the time the study was carried out. Ukpai and Ugwu, (2003) had earlier observed high prevalence rates for hookworm during the rainy season.

The relatively low prevalence rate of 5% of *Trichuris trichiura* was in contrast to observations made by Scolari *et al.*, (2000). But Bundy, (1986) had earlier observed that *T. trichiura* was typically an urban parasite.

The study indicated that prevalence of intestinal helminthes infection was higher in Orogodo Primary school 31.1% and Owanta 28.6%. These are public schools. In these schools, it was observed that personal hygiene was low and sanitation facilities were lacking. These could propagate the infections. Staff Model primary school and Hedson primary school though better-sanitized, recorded prevalence rates of 22.4% and 22.2% respectively. This could be attributed to the whole urban areas at risk for infection. The result (Table 6) showed further that Hedson primary school that had constant water supply had the lowest prevalence rate.

Prevalence of intestinal helminthes infection varied according to access to better facilities such as water supply and excreta disposal. Source of water was significantly associated with the occurrence of intestinal helminthes among subjects. The sources of water recorded in this study include; rainwater in concrete tanks, boreholes and public taps (pipe borne water). A fewer number obtained water from boreholes only. Most of these sources of water are not protected and are subject to contamination by cysts and ova of parasitic worms. The rate of infection observed among pupils who used borehole water was (14.9%). The possible sources of contamination could be from unwashed hands of children playing in the soil, they could in the process of fetching water contaminate the water container, handles of tap and the water itself. Preserved water in concrete tanks is stored over a long period of time and is exposed to series of contaminations from the environment. This could be responsible for the significant relationship between source of water and hookworm infection.

The age groups 5-7, 8-10 and 11-13 years recorded prevalence of 26.7%, 33.3% and 18.4% (Table 3) respectively for the three observed helminthes. The intestinal helminthes prevalence among this age group indicates a common pattern of behaviour and susceptibility for the age groups. Pupils within these age groups probably spend more time playing where they often come in contact with the soil. They also eat indiscriminately mostly with unwashed hands. However, the decrease in the prevalence of infection observed among children of between 11-13 years of age could be attributed to the fact that children of this age group are becoming more conscious of their personal hygiene. A similar trend was observed in Abia State by Ukpai and Ugwu, (2003).

The significantly higher prevalence of intestinal helminthes among males could be attributed to their activities such as playing football, which may predispose them to infections from the soil. Most times these activities are carried out barefooted. This could account for the high significant prevalence rate in males (34.4%) than their female counterparts (18.2%) for hookworm infection.

The high prevalence of infection observed among children who defecated in bushes (46.1%), hidden corners and sometimes poorly dug out pit latrines (32% and 29.5%) is expected if one considers the fact that their environment provides conducive local ecology for these helminthes to thrive. The lower prevalence of helminthes infections (19.7%) observed among water closet users may be attributed to the fact that the schools attended by these pupils and their homes lack toilet facilities and as such pupils would prefer to defecate in nearby bushes and farms around the school and thus infected from these areas, and also spread the parasites among those who visited those bushes and farms.

CONCLUSION/RECOMMENDATIONS

The considerable prevalence of intestinal helminthes in pupils of schools examined give vivid reflection of the poor state of hygiene in these schools therefore better sanitary facilities should be put in place in these schools.

Provisions should be made for adequate child friendly toilets, as this will reduce the amount of excreta found in the environment of these schools. A good source of water should be provided for the pupils at all times. The environments of these schools should be kept clean at all times. Screening of food handlers and vendors in these school should not be overlooked; and should be given necessary attention by the school management, moreover there should health education on how to maintain good personal hygiene both in school and at home such as regular washing of hands with soap and clean water especially after defecation and wearing of footwear could go a long way in preventing worm infestations.

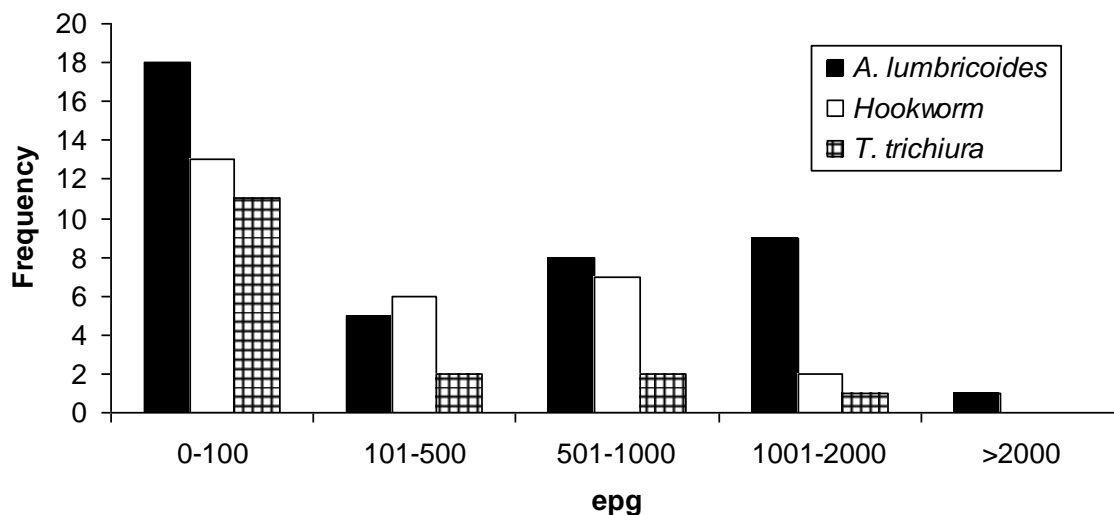


Fig 1 Frequency distribution of *A. lumbricoides* , hookworm and *T. trichiura* infections

TABLE 1: General Prevalence of Intestinal Helminthes Infection among Four Primary Schools in Agbor Metropolis

Name of Primary School	Total Number examined	<i>A. lumbricoides</i> Number Infected	Hookworm Number Infected	<i>T. trichiura</i> Number Infected	Total Number infected
Orogodo	90	15(16.7)	8 (8.9)	5 (5.6)	28 (31.1)
Staff Model	76	8(10.5)	7 (9.2)	2 (2.6)	17 (22.4)
Owanta	84	11 (13.1)	9 (10.7)	4 (4.8)	24 (28.6)
Hedson	72	7 (9.7)	4(5.6)	5 (6.9)	16 (22.2)
Total	322	41 (12.7)	28(8.7)	16 (5.0)	85 (26.4)

Percentages in parenthesis

TABLE 2: Prevalence of Intestinal Helminthes By Sex

Sex	Total Number examined	<i>A. lumbricoides</i> Number Infected	Hookworm Number Infected	<i>T. trichura</i> Number Infected	Total Number infected
Male	163	25 (15.3)	21 (12.9)	10 (6.1)	56 (34.4)
Female	159	16 (10.1)	7(4.4)	6 (3.8)	29 (18.2)
Total	322	41 (12.7)	28(8.7)*	16 (5.0)	85 (26.4)*

Percentages in parenthesis *P<0.05

TABLE 3: Prevalence of Intestinal Helminthes Infection by Age

Age	Total Number examined	<i>A. lumbricoides</i> Number Infected	Hookworm Number Infected	<i>T. trichura</i> Number Infected	Total Number infected
5-7	105	12(11.4)	7 (6.7)	9 (8.6)	28 (26.7)
8-10	114	22(19.3)	12 (10.5)	4 (3.5)	38 (33.3)
11-13	103	7 (6.8)	9 (8.7)	3 (2.9)	19 (18.4)
Total	322	41 (12.7)	28(8.7)	16 (5.0)	85 (26.4)

Numbers in parenthesis indicate percentages

TABLE 4: Prevalence of Intestinal Helminthes Infection by Toilet Type

Toilet Type	Total Number examined	<i>A. lumbricoides</i> Number Infected	Hookworm Number Infected	<i>T. trichura</i> Number Infected	Total Number infected
Water closet	137	14(10.2)	8 (5.8)	5 (3.6)	27 (19.7)
Pit latrine	122	16(13.1)	12 (9.8)	8 (6.6)	36 (29.5)
Pit latrine/nearby bush	50	9 (18.0)	6 (12.0)	1 (2.0)	16 (32.0)
Nearby bush	13	2 (15.4)	2(15.4)	2(15.4)	6 (46.1)
Total	322	41 (12.7)	28(8.7)*	16 (5.0)	85 (26.4)*

Percentages in parenthesis

* P<0.05

TABLE 5: Prevalence of Intestinal Helminthes Infection by Source of Drinking Water

Source of Water	Total Number examined	<i>A. lumbricoides</i> Number Infected	Hookworm Number Infected	<i>T. trichura</i> Number Infected	Total Number infected
Borehole	67	5(7.5)	3 (4.5)	2 (3.0)	10 (14.9)
Shallow well	87	14(16.1)	9 (10.3)	4 (4.6)	27 (31.0)
Pipeborne/Shallow well	68	12 (17.6)	6 (8.8)	5 (7.4)	23 (33.8)
Borehole/Shallow well	100	10 (10.0)	10(10)	5(5.0)	25 (25)
Total	322	41 (12.7)	28(8.7)*	16 (5.0)	85 (26.4)*

Percentages in parenthesis

* P<0.05

TABLE6: Description of Sanitation Conditions in the Four Primary Schools

Characteristics	Name of Primary schools			
	Orogodo	Staff Model	Owanta	Hedson
Source of water	Rainwater in concrete tanks	Borehole	Rainwater in concrete tanks	Borehole
Condition of Water supply	Not Constant	Constant	Not Constant	Constant
Latrine Facility	None	Water Closet	Pit latrine	Water closet/Pit
Sanitation of Latrine Facility	occasional	occasional	occasional	occasional
Available soaps on laboratories	none	None	None	none
Presence of Garbage piles around school	Present	Present	Absent	Absent
Screening of Food handlers	Yes	Yes	Yes	Yes

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**ANTIBIOTICS SUSCEPTIBILITY PATTERNS OF BACTERIAL SPECIES
ISOLATED FROM FRESH SEA FISHES SOLD IN MARKETS IN WARRI, DELTA
STATE NIGERIA.**

Owhe-Ureghe, U.B.

Department of Microbiology, Delta State University, Abraka

Abstract

Five fresh sea fishes (*Tilapia sp*, *Chrysichthys nigroditatus*, *Citharinus sp*, *Tilapia guineensis*, and *Lates niloticus*) bought from the main market and makava market in Warri Delta State Nigeria were investigated for their bacterial flora using standard conventional plating techniques. The total aerobic viable counts ranges from 7.5×10^6 CFU/mL for *Lates niloticus* to 7.0×10^7 CFU/mL for *Citharinus sp*; while the mean coliform counts range from 3.6×10^5 CFU/mL for *Citharinus sp* to 4.4×10^6 CFU/mL for *Tilapia guineensis*. Eight bacterial species were isolated; these include; *Bacillus cereus*, *Vibrio cholerae*, *Vibrio parahaemolyticus*, *Aeromonas hydrophila*, *Escherichia coli*, *Salmonella sp*, *Pseudomonas aeruginosa* and *Shigella dysenteriae*. The sensitivity profile of the isolates using some antibiotics battery shows that *Vibrio cholerae* and *Aeromonas hydrophila* were sensitive to all the antibiotics, while other bacterial isolates elaborated varying degrees of sensitivity and resistance. The diameter of the zones of inhibition for sensitive isolates range from 16-27 mm, while that for the resistant isolates ranged from 0-13mm. The result also shows that *Pseudomonas aeruginosa* was resistant to 8 out of the 15 antibiotics used in this study, while the remaining bacterial isolates were resistant to 1-4 antibiotics. The occurrence of pathogenic bacterial species with multiple drug resistance in sea fishes called for the proper pre-treatment of human, animal and domestic wastes before being dumped into the sea and fresh water environment, in order to reduce the possible contamination of this essential food resource (fish) with the consequent health implications.

Introduction

In the world today, there is a reemergence of the consumption of fish due to the new awareness about its low cholesterol, fat and good quality of animal protein. Fish is an essential food item in the diet of many people in West Africa. It provides an average of 35% of the total animal protein intake (FAO, 1978). Among coastal and riverian people, fish consumption is higher and contributes more than 50% of the animal protein in their diet e.g 70% in Sierra Leone, 80% in Ghana (Halliday, 1986). Nerquaye-Tetteh, (1986) reported that much of the fish consumed in West Africa region consist of cheap sea species, such as Sardinella, Bonga, Mackerel, Horse Mackerels, Anchovies and Tilapia.

In Nigeria, the populace depend more on sea fishes as fresh water species are more expensive and are bought by the elites in the society. This is an important feature of the domestic demand for fish in the sub region where income is generally low. In many Nigerian communities, preference for meat type is influence by religious beliefs and taboos, contrary to fish which is generally accepted by all.

Fish as a food is indispensable to the maintenance of good healthy living because of its high protein content; however, it can also be responsible for ill health (Adams and Moss, 1999). There are intrinsic and extrinsic hazards associated with fish consumption, but the hazard of primary concern is that of microbial origin. Hazards of other origin can be controlled during the processing but that of microbial origin is most difficult to control due to poor personal hygienic practices of the fish handlers and ignorance of the sources of contamination. Diseases associated

with the consumption of fish and fish products are not commonly reported in Nigeria due to lack of monitoring and non-attendance of patients to government hospitals as a result of lack of fund to pay for hospital bills.

Sea and fresh water fish business in the environs of Warri, Delta State Nigeria is very lucrative as many people patronize the sellers. Fish consumption is not affected by religion and is used as a special delicacy in many ceremonies such as marriages, burial and birthday parties. A proportion of the sea fishes and fresh water fishes sold in Warri and its environs are fresh (unfrozen) types in contrast to the frozen sea fishes sold in the hinterland.

Several gram positive (*Clostridium botulinum*, *Staphylococcus aureus*, *Bacillus cereus*, *Streptococcus sp.*), and gram negative bacterial species (*Escherichia coli*, *Shigella sp.*, *Salmonella sp.*, *Pseudomonas aeruginosa*, *Vibrio sp.*, *Aeromonas*, *Pasteurella multocida*) have been associated with fishes harvested from fresh water and marine environments (Lewis, 1975; Nair *et al.*, 1975; Sarkar *et al.*, 1985; Nizan and Hammerschida, 1993; Kori-siakpere, and Owhe-Ureghe 2001). Report by FAO fisheries department indicated that most bacterial species isolated as bacterial flora are implicated as aetiologic agents of fish diseases (Sakata and Hattori, 1988) a finding which may favour a concept of fish-borne human zoonotic infections.

It has been suggested that the bacterial flora of fishes is a reflection of the aquatic environment from which they were harvested (Showan and Hobbs, 1967). This has also been corroborated by Kori-siakpere and Owhe-Ureghe (2001) who reported the bacterial flora of *Channa obscura* from Ilushi river, Edo State Nigeria.

The health implications of the consumption of unwholesome food and water are centered on the contamination of these materials with bacterial species and other microbes, (Adams and Moss, 1999). In the developed and developing countries, the consequences of food borne illness, like diarrhoea diseases are enormous, because it is a major cause of morbidity and mortality, particularly among children. An estimated billion (10^9) episodes occur each year and nearly 5 million children under age of 5 die as a result.

Krumperman (1983) proposed that antibiotic resistance patterns of *E. coli* can be used as “fingerprints” to determine the source of the faecal contamination and that organisms obtained from the environment shows varying degrees of susceptibility and resistance to antibiotics. The paucity of information on the bacterial flora of fishes and their antibiotic sensitivity patterns in this locality prompted me to determine the bacterial flora and antibiotic sensitivity patterns of these bacterial flora of five fresh sea fishes sold in some markets in Warri, Delta State, Nigeria; to some commonly used antibiotics; Compare the antibiotics susceptibility patterns of isolates from the different species of fishes investigated.

Materials and Methods

Materials:

Materials used in this study includes, test tubes, conical flask, Pipettes, Glass slides, Petri dishes, non-absorbent cotton wool, Bunsen burner, Metler balance, chemical balance, Incubator, Autoclave, Refrigerator, Dissecting board and set, Mortar and Pestle, Whatman no 1 filter paper and Antibiotics sensitivity disks.

Reagents and Media

Grams reagents, Lead acetate paper, 70% alcohol, Methylated spirit, MacConkey agar, Nutrient agar, Nutrient broth, Urea agar, Citrate agar, Thiosulphate citrate bile salt sucrose agar, Oxidase strip reagent, hydrogen peroxide, indole reagents and 7 different sugars.

Samples Collection

The five sea fishes, *Tilapia sp.*, *Chrysichthys nigroditatus*, *Citharinus sp.*, *Tilapia guineensis*, and *Lates niloticus* weighting 50-120g, were bought from Warri main market and Makava market in Warri, Delta State, Nigeria. These were transported to the microbiology laboratory, Delta State University, Abraka in ice packed container for analysis, within 1hr. The photographs of the fishes are presented in Fig. 1.

FIG.1 THE FIVE FRESH SEA FISHES



Tilapia sp.



Tilapia guineensis



Citharinus sp.



Chrysichthys nigrodigitatus



Lates niloticus

Microbiological Analysis: The fishes were weighed with a chemical balance and disinfected externally with 70% ethanol. Each was then cut opened from the anus to the mouth region and the gut removed under aseptic condition. Approximately 1.0g of the gut was homogenized in 9mL of normal saline using a sterile Mortar and Pestle. The homogenate was then transferred into a sterile Pyrex glass boiling tube and further serially diluted ten-fold in normal saline. The diluted samples were then pour plated into freshly prepared sterile molten Nutrient agar, MacConkey agar, and Thiosulphate citrate bile salt sucrose agar to determine the total aerobic viable, coliform, and vibrio counts respectively as described by Harrigan and McChance (1982). The arithmetic mean of the counts at a chosen dilution was used to calculate the microbial population in the original samples.

Typical coliform, vibrio and viable colonies were randomly sub cultured onto sterile media and tested for their Gram reaction, motility, H₂S production, formation of spores, citrate utilization, carbohydrate fermentation, ability to produce indole, catalase, oxidase and urease enzymes. The various isolates were subsequently identified with the scheme of Cowan and Steel, (1985) and recorded in Tables 2 and 3.

Antibiotics Sensitivity Testing

About 1mL of overnight cultures of the isolates in nutrient broth was flooded on sterile nutrient agar plates. Antibiotic sensitivity discs of commonly used antibiotics were then placed on the surface of the seeded agar plates. These were allowed to stabilize and then incubated of 37°C for 24hr. The zones of inhibition of the different antibiotics were measured and recorded in Table 4.

RESULTS

The result of bacterial burden of the five sea fishes recorded in Table 1 shows that the mean total aerobic viable counts ranged from 7.5×10^6 CFU/mL for *Lates niloticus* to 7.0×10^7 CFU/mL for *Tilapia guineensis*. The mean coliform counts obtained ranged from 3.6×10^5 CFU/mL for *Chrysichthys nigroditatus* to 4.4×10^6 CFU/mL for *Tilapia guineensis*; while the mean *Vibrio* counts of the fishes ranged from 2.2×10^2 CFU/mL for *Citharinus sp* to 2.5×10^2 CFU/mL for *Tilapia guineensis*.

In all, 8 bacterial species were isolated (Tables 2) from the 5 types of fresh five sea fishes investigated. They include *Bacillus cereus*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella* sp, *Shigella dysenteriae*, *Vibrio cholerae*, *Vibrio parahaemolyticus* and *Aeromonas hydrophila*. Tables 3 show the comprehensive list of all the bacteria species isolated. *Salmonella* sp, *Aeromonas hydrophila*, and *Pseudomonas aeruginosa* were obtained, from all the five fresh sea fishes while *Bacillus cereus* was isolated from *Tilapia* sp, *Chrysichthys nigroditalus*, *Tilapia guineensis* and *Lates niloticus*. *Vibrio parahaemolyticus* was obtained from all the fishes except *Tilapia* sp, while *Shigella dysenteriae* was isolated from 3 fishes except *Tilapia* sp and *Tilapia guineensis*.

The diameter of the zones of inhibition (Tables 4) of the sensitive isolates ranged from 16 - 24mm, while that for the resistant isolates ranged from 0-13mm. The result (Tables 5) also shows that *Pseudomonas aeruginosa* was resistant to 8 out of the 15 antibiotics used in this study, while the remaining bacterial isolates were resistant to 1 - 4 antibiotics.

Table 1: Bacterial load of five fresh sea fishes (CFU/mL)

	TS	CN	CS	TG	LN
Mean total viable counts	8.1×10^6	8.3×10^6	7.0×10^7	8.0×10^6	7.5×10^6
Mean coliform counts	4.5×10^5	5.1×10^5	3.6×10^5	4.4×10^6	5.0×10^5
Mean vibrio counts	3.1×10^2	2.5×10^2	2.2×10^2	2.5×10^3	3.3×10^2

Key: TS = *Tilapia* sp, CN = *Chrysichthys nigroditatus*, CS = *Citharinus* sp
TG = *Tilapia guineensis*, LN = *Lates niloticus*

Table 2: Biochemical Characterization of the Isolates

Gram stain	Shape	Motility	Catalase	Oxidase	Urease	Indole	Spore formation	H ₂ S production	Citrate utilization	Glucose	Lactose	Mannitol	Sucrose	Sorbitol	Raffinose	Arabinose	Suspected Isolates
+	R	+	+	NT	NT	-	+	NT	+	+	-	+	+	+	+	+	<i>Bacillus cereus</i>
-	R	+	+	-	-	+	NT	-	+	+	+	+	+	+	+	+	<i>Escherichia coli</i>
-	R	+	+	+	+	-	NT	-	+	+	+	+	-	+	+	+	<i>Pseudomonas aeruginosa</i>
-	R	+	+	-	-	-	NT	+	-	+	-	-	-	+	-	+	<i>Salmonella species</i>
-	R	-	+	-	-	+	NT	-	-	+	-	+	-	+	-	+	<i>Shigella dysenteriae</i>
-	CR	+	-	+	-	+	NT	-	-	+	-	+	+	-	-	-	<i>Vibrio cholerae</i>
-	CR	+	-	+	-	+	NT	-	-	+	-	+	-	-	-	-	<i>Vibrio parahaemolyticus</i>
-	R	+	-	+	-	+	NT	-	+	+	+	-	+	-	-	+	<i>Aeromonas hydrophila</i>

Table 3: Bacterial isolates obtained from five fresh sea fishes

	Fish type	Bacteria Obtained
1	<i>Tilapia sp</i>	<i>Escherichia coli</i> , <i>Vibrio cholerae</i> , <i>Bacillus cereus</i> , <i>Salmonella sp</i> , <i>Aeromonas hydrophila</i> , <i>Pseudomonas aeruginosa</i> .
2	<i>Chrysichthys nigroditatus</i>	<i>Bacillus cereus</i> , <i>Vibrio cholerae</i> , <i>Vibrio parahaemolyticus</i> , <i>Aeromonas hydrophila</i> , <i>Escherichia coli</i> , <i>Salmonella sp</i> , <i>Pseudomonas aeruginosa</i> , <i>Shigella dysenteriae</i>
3	<i>Citharinus sp</i>	<i>Vibrio cholerae</i> , <i>Shigella dysenteriae</i> , <i>Escherichia coli</i> , <i>Salmonella sp</i> , <i>Aeromonas hydrophila</i> , <i>Pseudomonas aeruginosa</i> , <i>Vibrio parahaemolyticus</i>
4	<i>Tilapia guineensis</i>	<i>Vibrio cholerae</i> , <i>Vibrio parahaemolyticus</i> , <i>Bacillus cereus</i> , <i>Aeromonas hydrophila</i> , <i>Escherichia coli</i> , <i>Salmonella sp</i> , <i>Pseudomonas aeruginosa</i>
5	<i>Lates niloticus</i>	<i>Escherichia coli</i> , <i>Shigella dysenteriae</i> , <i>Vibrio cholerae</i> , <i>Salmonella sp</i> , <i>Bacillus cereus</i> , <i>Vibrio parahaemolyticus</i> , <i>Aeromonas hydrophila</i> , <i>Pseudomonas aeruginosa</i>

Table 4: Antibiotics Susceptibility pattern of the various bacterial isolates

Isolates	Zones of inhibition of Antibiotics (mm)														
	Chloramphenicol 25µg	Cefurazone 10 µg	Nitrofurantoin 15 µg	Gentamicin 10 µg	Ampicillin 10 µg	Augmentin 10 µg	Nalidixic acid 5 µg	Amoxycillin 15 µg	Peflaxacin 25 µg	Tetracycline 10 µg	Ofloxacin 15 µg	Cotrimoxazole 30 µg	Ciprofloxacin 30 µg	Erythromycin 10 µg	Ceporex 5 µg
<i>Escherichia coli</i>	23	13	25	26	11	26	27	25	23	23	25	25	23	25	23
<i>Pseudomonas aeruginosa</i>	0	0	10	0	9	21	20	20	11	0	24	24	20	19	11
<i>Salmonella species</i>	9	22	25	27	22	28	25	23	24	9	23	26	22	9	10
<i>Bacillus species</i>	17	15	9	20	8	9	20	16	17	16	10	18	16	19	17
<i>Shigella species</i>	20	21	24	22	8	9	22	26	20	22	21	10	22	23	21
<i>Vibrio cholera</i>	25	22	21	20	23	26	24	25	21	20	24	20	22	21	23
<i>Vibrio parahaemolyticus</i>	20	23	21	24	12	23	25	24	26	24	20	21	25	20	20
<i>Aeromonas hydrophila</i>	19	18	20	19	21	22	18	20	21	21	20	19	20	20	20

Table 5: The antibiogram of the bacterial isolates

Isolates	Antibiotics Zones of inhibition (mm)														
	Chloramphenicol 25 µg	Cefuroxime 10 µg	Nitrofurantoin 15 µg	Gentamicin 10 µg	Ampicillin 10 µg	Augmentin 10 µg	Nalidixic acid 5 µg	Amoxycillin 15 µg	Pefloxacin 25 µg	Tetracycline 10 µg	Ofloxacin 15 µg	Cotrimoxazole 30 µg	Ciprofloxacin 30 µg	Erythromycin 10 µg	Ceporex 5 µg
<i>Escherichia coli</i>	S	R	S	S	R	S	S	S	S	S	S	S	S	S	S
<i>Pseudomonas aeruginosa</i>	R	R	R	R	R	S	S	S	R	R	S	S	S	S	R
<i>Salmonella species</i>	R	S	S	S	S	S	S	S	S	R	S	S	S	R	R
<i>Bacillus species</i>	S	S	R	S	R	R	S	S	S	S	S	S	S	S	S
<i>Shigella species</i>	S	S	S	S	R	R	S	S	S	S	S	R	S	S	S
<i>Vibrio cholera</i>	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S
<i>Vibrio parahaemolyticus</i>	S	S	S	S	R	S	S	S	S	S	S	S	S	S	S
<i>Aeromonas hydrophila</i>	S	S	S	S	S	S	S	S	S	S	S	S	S	S	S

KEY:

R = Resistant

S = Sensitive

Discussion

Seafood has the potential to pose a wide spectrum of public health problems from harmful bacteria species through contamination during distribution from the point of harvest to final preparation. The result of this study (Tables 2 and 3) shows 6 - 8 Gram positive and Gram negative bacteria species associated with the gastrointestinal tract of the five sea fishes, these are related to earlier report by Sarkar *et al.*, (1985) from similar marine environment. Many of these organisms could have found their way into the water body through contamination from untreated domestic, animal and human wastes that are usually dumped into the marine water of Warri. This is supported by the report of Sarkar *et al.*, (1985) and Kori-siakpere and Owhe-Ureghe (2001), that the microbial load of sea and fresh water fishes is a reflection of the aquatic environment, the diets of the fish and the physio- chemical parameters of the environment from which they were harvested. One obvious implication of this school of thought is that the microbiological quality of the waters affects mobile or migratory species as well as sedentary shell fish. Contamination by enteric bacteria in polluted harvest area is sporadic and difficult to control as the result of this study tend to suggests. Apart from typical marine bacterial species like, *Vibrio cholerae*, *V. parahaemolyticus* and *Aeromonas hydrophila*, other isolates were enteric related.

The antibiotics susceptibility studies on the bacterial isolates obtained in this investigation shows that (Table 4 and 5), all were sensitive to Nalidixic acid, Amoxycillin, Ofloxacin and Ciprofloxacin, while they show varying degrees of sensitivities and resistance to other antibiotic used in this investigation. Apart from *Pseudomonas aeruginosa* that was resistant to 8 out of the 15 antibiotics used in this study. Out of the 8 bacterial species isolated, 6 (75%) were resistant to 1 - 4 antibiotics, while only 2 (25%) (*Vibrio cholerae* and *Aeromonas hydrophila*) were sensitive to all antibiotic used in this study. Multiple drug resistance phenomenon observed in this study due to *P. aeruginosa* and more than 50% of the bacterial isolates have also been reported in this Nigerian environment (Owhe-Ureghe *et al.*, 2000) and other parts of the world (Watkins and Cabelli, 1985). The result further showed that the *Vibrio sp* and *Aeromonas hydrophila* which are typical marine organisms were either 100% sensitive to all the antibiotics or resistant only to one antibiotic

Conclusion

The occurrence of pathogenic bacterial speaks with multiple drug resistance in sea fishes called for the proper pre-treatment of human, animal, and domestic waste before being dumped into the sea and fresh water environment, in order to reduce the possible contamination of this essential food resource (fish) with the consequent health implication. Furthermore, it is advisable to subject fish and fish products to adequate cooking or adequate heat exposure during processing so that these products could be hygienically safe for human consumption.

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THE IMPACT OF ENVIRONMENT ON SOME AQUATIC MACROPHYTES COLLECTED FROM SOME BRACKISH AND FRESH WATER RIVERS IN DELTA STATE, NIGERIA.

Noyo E. Edema

Department of Botany, Delta State University, Abraka.
E-mail: drangeledema@yahoo.com; Phone: 08028530761

Abstract

A preliminary investigation of the impact of environment on macrophytes in some brackish and fresh water rivers in Delta State were carried out using chlorophyll content and catalase activity as parameters. The sampled areas include River Ethiope and Uton creek (Sapele Local Government Area), Ubeji and Ugboiwangue (Warri South Local Government Area) and Otorho and Abraka river (Ethiope East Local Government Area). The macrophytes encountered include: *Eichhornia crassipes*, *Azolla africana*, *Salvinia nymhellula*, *Nymphaea lotus*, *Ceratophyllum demersum* and *Pistia stratiotes*. These species are free floating, except *Nymphaea lotus* which is an emergent floating macrophyte. Uton, Ubeji and Ugboiwangue are brackish, while River Ethiope, (Sapele), Abraka and Otorho river (Ethiope East L.G.A) are freshwater habitats. Sapele and Warri are within the oil producing area of Delta State. Otorho and Abraka rivers (control) are not within the oil producing area of Delta State. And all the macrophytes encountered during the sampling period were found in River Ethiope. This shows that macrophytes diversity was highest in River Ethiope. Otun creek had the second highest species diversity. Only one macrophyte (*Eichhornia crassipes*) was encountered in Ubeji and Ugboiwangue. This may be attributed to the brackish nature of the water bodies as compared to River Ethiope which is a fresh water body. The results indicated that chlorophyll content and catalase activity were more in *Eichhornia crassipes* collected from River Ethiope than in other rivers. The values recorded were $1.60 \pm 0.12 \text{ mg/g}^{-1} \text{ freshwt}$ and $25.60 \pm 0.12 \text{ mg/freshwt}$ chlorophyll content and catalase activity respectively. While *Nymphaea lotus* and *Pistia* species collected from River Ethiope and Otorho river had the least values of $10.50 \pm 0.13 \text{ mg/fresh wt}$ and $0.37 \pm 0.04 \text{ mg/g}^{-1} \text{ freshwt}$ for catalase activity and chlorophyll content respectively. The differences recorded may be attributed to differences in the nutrient level, the presence of toxic substances or water stress.

INTRODUCTION

Aquatic macrophytes are plants which germinate and grow in water and are large enough to be seen with unaided eyes. They grow in periodical or permanently submerged habitats and whose growth is favoured by water logging or when submerged (Lavania *et al.*, 1990). Aquatic macrophytes are found in all bodies of water such as flowing, standing, fresh and brackish environments. Macrophytes are not only flowering plants but also include ferns, bryophytes and algae (Langhans *et al.*, 2008)

Macrophytes constitute to the general fitness and diversity of a healthy aquatic ecosystem by acting as indicators of water quality and aiding in nutrient cycling (Flint and Madsen, 1995). Aquatic macrophytes are important components of many water courses, providing structures and habitat for fish and invertebrates offering protection against currents and predators and forming a substrate for the

deposition of eggs. As primary producers, macrophytes present an important food resources and they also play a significant role in the oxygen balance and nutrient cycle of many water courses (Langhans *et al.*, 2008) Macrophytes help in removing suspension particles and nutrients from the water column (Madsen *et al.*, 1996).

The effects of wave action or sediment induced turbidity can prevent the growth of submerged macrophytes (Kantrud, 1990). Light availability is considered to be the primary factor limiting the growth of submerged macrophytes (Scheffer, 1998). Aquatic plants grow in an environment that is often poor in mineral nutrients, since they can absorb and store large quantities of nutrients for later use. Aquatic plants are sensitive to changes (increase) in nutrient concentration and organic pollutants. The composition of macrophyte species in water body makes it difficult to draw conclusion about the local chemical and physical conditions. Species that prefer low nutrient concentrations have become much less prevalent. Concentrations of some mineral nutrients in plants, most notably micronutrients such as iron and copper exceed the level in water by 1,000 to 1,000,000 times. Regular addition of mineral nutrients, particularly iron are therefore essential for the sustained growth of aquatic plants in aquarium (Watzel, 2001).

Based on the life forms, growth and the plant position with respect to water surface they can be classified into four main types – embarkment (e.g *Calogonium mucunoids*) which is rooted, Submerged (e.g *Ceratophyllum demersus*) rooted to the bottom completely and under water, Emergent (e.g *Fuirena umbellata*) rooted in water with some or all the leaves and stem as arial and floating which are either attached or free floating e.g *Nymphaea lotus* and *Azolla africana*

Hydrogen peroxide is a harmful by-product of many normal metabolic processes. To prevent damage it must be quickly converted into other, less dangerous substances. Therefore, catalase which are found in many plants and animals (Srinivasa *et al.*, 2003).is frequently used by cells to rapidly catalyse the decomposition of hydrogen peroxide into less reactive gaseous oxygen and water (Amo *et al.*, 2002).

This study is aimed at determining the status of the biological properties of aquatic macrophytes from some rivers in Delta State, Nigeria

MATERIALS AND METHODS

STUDY AREA

The study areas include River Ethiope and Otun creek (Sapele Local Government Area), Ugbuwangue and Ubeji (Warri South Local Government Area) Otorho (Abraka inland) and Abraka river (Ethiope East Local Government Area) of Delta State. Sapele and Warri are within the oil producing area of Delta State, while the Abraka river and Otorho (were the control) are not within the oil producing area of Delta State. These towns are situated between latitude $5^{\circ}5^{\prime}$ and $6^{\circ}30^{\prime}$ N, latitude $5^{\circ}5^{\prime}$ and $6^{\circ}15^{\prime}$ E of the equator.

COLLECTION OF PLANT SAMPLES

Plant samples were collected by hand into plastic bags and marked with field numbers. These were transported to the laboratory and kept in aquarium until required for analysis.

DETERMINATION OF CHLOROPHYLL

One gramme of fresh leaf was placed in a ceramic mortar and 25ml of 80% acetone was added. Grinding was done until the chlorophyll was released. To The extract was filtered through Whatman No.1 filter paper and subsequently used for chlorophyll determination according to the method of Holden (1976).

DETERMINATION OF CATALASE ACTIVITY

Catalase activity was assessed by measuring the volume of gas (volume of oxygen) released by the macrophytes collected. Determination of Catalase activity was as modified by Moore (1974).

TABLE 1: AQUATIC MACROPHYTES ENCOUNTERED

S/N.	LOCATION	COMMON NAME	BOTANNICAL NAME	CLASSIFICATION	FAMILY
1.	River Ethiope	Water hyacinth	<i>E. crassipes</i>	Free floating	Pontederialeae
2.	River Ethiope	Water velvet	<i>A. africana</i>	Free floating	Azollaceae
3.	River Ethiope	Water lily	<i>N. lotus</i>	Attached but floating (Emergent)	Nymphaeaceae
4.	River Eithiope	Hornworts	<i>C. demersum</i>	Free floating (submergent)	Salviniaceae
5.	River Ethiope	Giant salvinia	<i>S. nymphellula</i>	Free floating	Araceae
6.	River Ethiope	Water lettuce	<i>P. stratiotes</i>	Free floating	Araceae
7.	Otun creek	Water lettuce	<i>P. stratiotes</i>	Free floating	
8.	Otun creek	Hornworts	<i>C. demersum</i>	Free floating (submergent)	Pontederialeae
9.	Otun creek	Water hyacinth	<i>E. crassipes</i>	Free floating	
10.	Otun creek	Water velvet	<i>A. africana</i>	Free floating	Azollaceae
11.	Otun creek	Water lily	<i>N. lotus</i>	Attached but floating (Emergent)	Nymphaeaceae
12.	Ugbuwange river	Water hyacinth	<i>E. crassipes</i>	Free floating	Pontederialeae
13.	Ubeji river	Water hyacinth	<i>E. crassipes</i>	Free floating	Pontederialeae
14.	Otorho	Water velvet	<i>A. africana</i>	Free floating	Azollaceae
15.	Otorho pond	Water lettuce	<i>P. stratiotes</i>	Free floating	Araceae
16.	Otorho	Giant salvinia	<i>S. nymphellula</i>	Free floating	Salviniaceae
17.	Otorho	Water lily	<i>N. lotus</i>	Attached but floating (Emergent)	Nymphaeaceae
18.	Abraka river	Water lily	<i>N. lotus</i>	Attached but floating (Emergeng)	Nymphaeaceae

STATISTICAL ANALYSIS

Mean and standard error were used for the statistical analysis (Ogbeibu, 2005).

Table 2: Mean values of catalase activity of the macrophytes encountered

LOCATION/SITE	Macrophytes	CATALASE ACTIVITY (mg/fresh wt)	CHLOROPHYLL CONTENT (mg/g-1 fresh wt)
Ubeji	<i>E. crassipes</i>	25.40±0.09	1.53±0.04
Ugbuwangue		20.80±0.06	1.53±0.06
River Ethiope		25.60±0.12	1.60±0.12
Otun creek		20.80±0.07	1.60±0.12
Abraka Inland		15.60±0.04	1.30±0.02
Abraka River		-	-
Ubeji	<i>N. lotus</i>	-	-
Ugbuwangue		-	-
River Ethiope		10.53±0.01	0.79±0.71
Otun creek		25.40±0.09	0.79±0.71
Abraka Inland		-	-
Abraka River		14.40±0.95	0.94±0.29
Ubeji	<i>C. demersum</i>	-	-
Ugbuwangue		-	-
River Ethiope		10.60±0.12	0.77±0.32
Otun creek		10.80±0.09	0.77±0.32
Abraka Inland		-	-
Abraka River		-	-
Ubeji	<i>A. africana</i>	-	-
Ugbuwangue		-	-
River Ethiope		20.70±0.09	0.55±0.12
Otun creek		20.50±0.09	0.53±0.10
Abraka Inland		20.50±0.06	0.56±0.06
Abraka River		-	-
Ubeji	<i>P. stratiotes</i>	-	-
Ugbuwangue		-	-
River Ethiope		15.30±0.09	0.53±0.10
Otun creek		20.53±0.07	0.37±0.04
Abraka Inland		-	-
Abraka River		-	-
Ubeji	<i>S. nymphyllula</i>	-	-
Ugbuwangue		-	-
River Ethiope		15.40±0.06	0.46±0.09
Otun creek		-	-
Abraka Inland		-	-
Abraka River		-	-

RESULTS AND DISCUSSION

The physiology of some macrophytes from some water bodies was conducted in order to determine the effect of the water environment on the biological properties of the macrophytes. Six different macrophytes were encountered. Five were the free floating type, while one was the emergent (attached but free floating) type. *Eichhornia crassipes* was encountered in all the locations (Table 1). And all the macrophytes encountered during the sampling period were found in River Ethiope. This shows that macrophytes diversity was highest in River Ethiope. Otun creek had the second highest species diversity. Only one macrophyte (*Eichhornia crassipes*) was encountered in Ubeji and Ugbuwangue. This may be attributed to the brackish nature of the water bodies as compared to River Ethiope which is a fresh water body. The difference in macrophyte diversity may also be due to the presence of ions or nutrient level of the water bodies.

The highest value of $25.60 \pm 0.06 \text{ mg/g}$ fresh wt of catalase was recorded for *Eichhornia species* in River Ethiope. While the least value of catalase activity was recorded for the same species from Otorho river (Table 2). Catalase activity was higher in *Nymphaea* species collected from Otun creek than River Ethiope (Table 2). *Pistia stratiotes* from Otorho river had the highest catalase activity compared to the other two locations (Table 2). Results show no much difference in catalase activity of *Cerarophyllum demersus* and *Azolla africana* of both sites. *Salvinia nymphellula* showed low catalase activity (Table 2). Carbon three (C_3) plants have been shown to have high catalase activity, while C_4 plants are known to have low catalase activity. Catalase existing in green plant tissues is important in glycolate metabolism, oxidation of glycolate to glyoxylate which results in the production of hydrogen peroxide (H_2O_2). The peroxide however is rapidly decomposed to water and molecular oxygen. The increase in catalase activity in the macrophytes was to enable the plants to cope with the increase level of hydrogen peroxide thereby, rapidly catalyzing the decomposition of H_2O_2 into less reactive gaseous oxygen and water. The increase catalase activity in plants collected from brackish water may due to the presence of macro elements such as calcium, magnesium etc. (Case and Madsen, 2004). There are some indication that salt stress can reduce condition of oxidative stress (Burdon *et al.*, 1996). Catalase activity increased in *Salvinia* and *Pistia* shoot and root tissues in response to salinity (NaCl) Upadhyay and Panda, 2005.

Eichhornia species collected from River Ethiope and Uton creek had the highest chlorophyll content of $1.60 \pm 0.12 \text{ mg/g}^{-1}$ freshwt, while Otorho had the least value of $1.30 \pm 0.02 \text{ mg/g}^{-1}$ freshwt. *Pistia* species encountered in River Ethiope recorded higher value of $0.53 \pm 0.10 \text{ mg/g}^{-1}$ freshwt when compared to $0.37 \pm 0.04 \text{ mg/g}^{-1}$ freshwt presented in Uton creek. Chlorophyll content of $0.46 \pm 0.09 \text{ mg/g}^{-1}$ freshwt was recorded for *Salvinia nymphellula*. Chlorophyll degradation is a symptom of damage caused by environmental pollution (Ghanima *et al.*, 1996). Chlorophyll cerotenoid ratio suffer as a result of different kinds of environmental stress (Ghanima *et al.*, 1996). Upadhyay and Panda, 2005 reported decrease in chlorophyll and cerotenoid contents with increase in NaCl (salinity) for *Salvinia* species.

This study is a preliminary investigation of the extend to which the aquatic macrophytes have been impacted by the water environment. There is the need to look at the physicochemical characteristics of the different water bodies in order to make a firm conclusion on the effect of the water bodies in Delta State environment on the aquatic macrophytes. Also, in subsequent studies the number of locations would be increased.

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PHYTOCHEMICAL SCREENING AND ANTIBACTERIAL ACTIVITIES OF EXTRACTS OF ALLIUM CEPA (ONION) PEELS

Owhe-Ureghe U. B. and Akpona E

Department of Microbiology Delta State University, Abraka, Nigeria.

Abstract

The antibacterial efficacy of cold water, hot water and ethanol extracts of *Allium cepa* (onion) peels against *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* was determined using the disc diffusion method. The results revealed that, growth of the test isolates were inhibited to varied degrees by the cold water and ethanol extracts. However, the isolates were predominantly resistant to the activity of the hot water extract. The zones of inhibition of the ethanol extract against *K. pneumoniae*, *P. aeruginosa* and *S.aureus* ranged between 3.0-6.0 mm, 9.0-27.0 mm and 3.0-14.0 mm respectively. Exposure to cold water extract resulted in zones of inhibition that ranged between 2.0-7.0 mm for *K. pneumoniae*, 7.0-15.0 mm for *P. aeruginosa* and 2.0-10.0 mm for *S. aureus*. Minimum inhibitory concentrations of cold water extracts obtained were 50 mg/ml on *S. aureus* and 100 mg/ml for both *K. pneumoniae* and *P.aeruginosa*.The hot water extract had no inhibitory activity on the three test isolates up to the highest concentration of 200 mg/ml. When *K. pneumoniae* was exposed to varied concentrations of ethanol extract, 100 mg/ml gave the minimum inhibitory concentration while 50 mg/ml was for both *Pseudomonas aeruginosa* and *Staphylococcus aureus*. Increase in the storage duration of the extracts had no significant effect on their inhibitory activities. Phytochemical screening of the various extracts revealed the presence of tannins, phenols, glycosides, alkaloids and flavonoids. The results of this study therefore, suggest the possibility of using extracts from onion peels as an antibacterial agent which would also help to curb the nuisance constituted by this hitherto waste product and thus, converting ‘waste to wealth’.

Keywords: Phytochemical, antibacterial, extract, *Allium cepa*.

INTRODUCTION

The practice of herbal medicine in modernized form is now gaining ground in Nigeria as there is increasing awareness of the potencies and efficacies of some indigenous plants (Nwangu, 1997).Thus, the use of herbal remedies is increasingly being incorporated into orthodox medical practice (Olukemi and Kandakai-Olukemi, 2004). Medicinal uses of these plants range from the administration of the plant roots, barks, stem, leaves and seeds to the use of extracts from the whole plant (Ogbulie *et al.*, 2004).

The search for new antimicrobial agents is associated with the problems of emergence of strains that are resistant to most present day antibiotics (Ibekwe *et al.*, 2000).Also, researchers have revealed that certain herbs/foods and their individual constituents perform in a fashion similar to modern drugs and sometimes better without the dreaded side effects.

This paper reports investigations into the antibacterial activities of the extracts of *Allium cepa* (onion) peels against some clinical isolates responsible for upper respiratory tract infections. The onion is one of the oldest cultivated vegetable in history. It consists of the herbaceous plant part and the edible bulb part. The antibacterial activities of the onion bulb on different microorganisms such as fungi, bacteria and viruses have been studied (Irkin and Korukluoglu, 2007; Ghahfaroklu *et al.*, 2003). Also, its phytochemical components have important applications in cancer chemoprevention, inhibition of aflatoxin synthesis, mycotoxin-induced toxicity and free-radical formation as well as prevention of Hepatitis A (Park and Pezzuto, 2002; Abdel and Aly, 2003).

This study was designed in view of the fact that there are still parts of the plant such as the peels whose medicinal values have not been ascertained.

MATERIALS AND METHODS

Test organisms

The test organisms were clinical isolates obtained from Eku Baptist Hospital, Eku, Delta State. They include; *K. pneumoniae*, *P. aeruginosa* and *S.aureus*. The isolates were sub-cultured on nutrient agar slants and preserved in the refrigerator at 4°C until required for the study. Confirmation of the identity of each isolate was done according to the method of Cowan and Steel, 1965.

Plant preparation and extraction

The plant material used was the peels of onion bulb. They were obtained from Abraka main market, Abraka, Delta State. The peels were sun-dried until constant weight was achieved and were then ground into powder form. The active compounds were extracted by soaking 200g of samples (powder form) into 600 ml of appropriate solvents for about 24h with intermittent shaking according to the method of Akujobi *et al.*, 2004. Water and Ethanol were used as extractants. Cold and hot water extraction were done. For hot water extraction, the sample and solvent were heated to boil for 120 min and then allowed to stand for 24 h. At the end of extraction duration, filtration using Whatman no 1 filter paper was done, the solvent was evaporated using rotary evaporator and the extracts stored for further analysis.

Antibacterial screening test

The sensitivity of the test organisms *K. pneumoniae*, *P.aeruginosa* and *S. aureus* to extracts of *A. cepa* peels were carried out using the agar diffusion method.

The respective bacterial inoculum was standardized by inoculating 5 ml of sterile nutrient broth with 5 colonies of 24 h culture of appropriate test organism. These were incubated at 37°C until turbidity reached that of 1.0% BaSO₄ solution. Each standardized inoculum was then swabbed inoculated onto freshly prepared Muller-Hinton agar plates and the excess fluid was allowed to dry for about 5min. Sterile discs made from filter paper (Whatman no 1), were then impregnated with 2 ml of the various concentrations of the extracts and aseptically placed on the agar. Incubation followed immediately at 37°C for 18-24h. The concentrations of the extracts used were 12.5 mg/l, 25 mg/l, 50 mg/l, 100 mg/l and 200 mg/l.

Zones of inhibition obtained were measured and recorded against corresponding concentrations.

Determination of minimum inhibitory concentration (MIC)

The test tube dilution method described by Ibekwe *et al.*, 2001 was used. The various concentrations of the respective extracts were dispensed in 2 ml amounts into sterile test tubes and then 5 ml of standardized inoculum were introduced into each and incubated at 37°C for 24h. The lowest concentration of the extract that inhibited the growth of the test organism was recorded as minimum inhibitory concentration.

Determination of the effect of storage duration on activity of extracts

The dried extracts obtained were wrapped in foil paper and kept in a clean and dry cabinet for duration of 60 days. However, at storage duration of 0, 7, 21 and 60 days, the extracts was re-constituted to prepare the

minimum inhibitory concentration obtained against each test isolate as described above. Sterile filter paper discs were then impregnated with 2 ml of this and aseptically placed on the surface of Muller- Hinton agar plates that have been seeded with appropriate standardized inoculum. Incubation followed immediately at 37°C for 18-24 h at the end of which zones of inhibition were recorded.

Phytochemical screening

The extracts obtained were screened for the presence of tannins, alkaloids, flavonoids, glycosides and phenols using methods adapted from AOAC, 1980.

RESULTS

The results of the antibacterial properties of the ethanolic, hot water and cold water extracts of onion peels on the test organisms are shown in Tables 1, 2 and 3, respectively. The water and ethanol extracts of onion peels showed activities against the three test isolates with the highest activity recorded against *P. aeruginosa* followed by *S. aureus* and the least against *K. pneumoniae*. It was however clear from the data obtained that the extractant affected the degree of the activities of the various extracts. The ethanolic extract (Table 1) gave the widest zones of inhibition ranging from 3.0 to 6.0 mm for *K. pneumoniae*, 9.0 to 27 mm for *P. aeruginosa* and 3.0 to 14 mm for *S. aureus*. The hot water extracts of onion peels as shown in Table 2 did not inhibit the growth of *K. pneumoniae* at all and generally, the zones of inhibition produced against *P. aeruginosa* and *S. aureus* were the lowest of the three extracts.

The minimum inhibitory concentration (MIC) of the extracts on the respective isolate is shown in Table 4. The hot water extract had no inhibitory activity on the three test isolates up to the highest concentration of 200 mg/ml. The MIC of the cold water extract was 100 mg/ml against both *K. pneumoniae* and *P. aeruginosa* and 50 mg/ml against *S. aureus*. When *K. pneumoniae* was exposed to ethanol extract, 100 mg/ml was the MIC while 50 mg/ml was for both *P. aeruginosa* and *S. aureus*.

The results of the effect of storage duration on extracts activities presented in Tables 5, 6 and 7 indicate that none of the extract lost its inhibitory activity with increase in storage duration within a 60 day period. Also, at 95% confidence limit there was no significant difference in activity.

The result of the Phytochemical screening as presented in Table 8 revealed the presence of tannins, flavonoids phenols, glycosides and alkaloids.

DISCUSSION

The result obtained in this study revealed that onion peels contain antimicrobial substances. This agrees with the reports of previous workers who have shown that many plants possess antimicrobial activities (Lewis, 1980; Zaria *et al.*, 1995; Ibekwe *et al.*, 2001; Akujobi *et al.*, 2004 and Akujobi *et al.*, 2006).

Data obtained showed that the various soluble extracts of onion peels had antibacterial properties. The widest zones of inhibition obtained were with *P. aeruginosa* followed by *S. aureus* and then *K. pneumoniae*. This difference in the zones of inhibition obtained may be directly related to the susceptibility of each test organism to the onion peel extracts. The factors responsible for the difference in susceptibility of test isolates probably are inherent. The presence of the capsule in *K. pneumoniae* might have conferred increased resistance to the test isolate. The higher sensitivity of *P. aeruginosa* to the various extracts than *S. aureus* might be due to the differences in their cell wall composition. Similar observations had been made by Akujobi *et al.*, 2006. The report of this author indicates that Gram negative isolates were more sensitive to various plant extracts than Gram positive isolates.

Furthermore, results obtained showed that ethanolic extracts gave the widest zones of inhibition to the three test isolates. This may be due to the fact that ethanol being an organic solvent was able to dissolve organic compounds better, thus, liberating the active components required for antibacterial activity. This also is in accordance with the observations of Ibekwe *et al.*, 2001; Akujobi *et al.*, 2006.

The hot water extract showed the least activity against the test organisms. This is an indication that the active components of the peels are heat-labile. It has been shown that the antibacterial components of spice plants are not thermally- stable.

Results of the phytochemical screening of the onion peel extracts revealed the presence of phytochemical compounds in the onion peels and thus provide evidence for the antibacterial activity of the sample. Storage of extracts for duration of 60 days did not lead to the loss of their potential suggesting its adequacy as an effective antibacterial agent that would not loose potency irrespective of storage within tested duration.

Finally, the use of *Allium cepa* peels as an antibacterial agent (alternative to orthodox medicine) would help to curb the nuisance constituted by this hitherto waste product and hence converting “waste to wealth”.

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Table 1. Sensitivity pattern of test isolates to ethanolic extract of *Allium cepa* peels

Concentration (mg/ml)	Diameter of zones of inhibition(mm)		
	<i>K. pneumoniae</i>	<i>P. aeruginosa</i>	<i>S. aureu s</i>
12.5	NI	NI	3.0
25	3.0	9.0	5.0
50	4.0	12.0	9.0
100	4.0	23.0	19
200	6.0	27.0	14

NI = No Inhibition

Table 2. Sensitivity pattern of test isolates to hot water extract of *Allium cepa* peels

Concentration(mg/ml)	Diameter of zones of inhibition(mm)		
	<i>K. pneumonia</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>
12.5	NI	NI	NI
25	NI	NI	NI
50	NI	NI	4.0
100	2.0	2.0	NI
200	NI	2.0	1.5

NI = No Inhibition

Table 3: sensitivity pattern of *K. pneumoniae*, *P. aeruginosa* and *S. aureus* to cold water extract of *Allium cepa* Peels

Concentration mg/ml	Diameter of zones of inhibition(mm)		
	<i>K. pneumoniae</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>
12.5	N.I	N.I	N.I
25	N.I	7.0	2.0
50	2.0	10.0	5.0
100	7.0	13.0	11.0
200	7.0	15.0	10.0

NI = No Inhibition

Table 4. Minimum inhibitory concentration (MIC) of various extracts on test isolates

Extract	Minimum inhibitory concentration (mg/ml)		
	<i>K. pneumoniae</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>
Cold water	100	100	50
Hot water	-	-	-
Ethanol	100	50	50

Table 5. Effect of storage duration on inhibitory activity of ethanolic extract on various test organisms

Storage duration (day)	Diameter of zones of inhibition (mm)		
	<i>K. pneumonia</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>
0	14	21	13
7	13	21	13
21	12	20	13
60	13	21	12

Table 6. Effect of Storage Duration on Inhibitory Activity of Hot water Extract on Various Test Organisms

Storage Duration (Day)	Diameter of zones of inhibition (mm)		
	<i>K. pneumonia</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>
0	NI	2.0	1.5
7	NI	1.0	1.0
21	NI	NI	NI
60	NI	NI	1.0

Note: NI = No Inhibition

Table 7. Effect of Storage Duration on Inhibitory Activity of Cold water Extract on Various Test Organisms

Storage duration (day)	Diameter of zones of inhibition (mm)		
	<i>K. pneumoniae</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>
0	7.0	13.0	11.0
7	7.0	11.0	11.0
21	8.0	12.0	11.0
60	7.0	12.0	11.0

Table 8. Phytochemical Components of the Various *Allium cepa* extracts

Phytochemical component	Ethanolic extract	Hot extract	water	Cold extract	water
Flavonoids	+	-			+
Alkaloids	+	-			+
Tannins	+	+			+
Glycosides	+	+			+
Phenols	+	+			+

A SOCIO-SCIENTIFIC ASSESSMENT OF CONSUMER EATING BEHAVIOUR IN RESTAURANTS OF DELTA STATE UNIVERSITY ABRAKA, NIGERIA.

Ehwarieme D.A., Oghenemowho E. and Ejechi B.O.

Department of Microbiology, Delta State University, Abraka.

Abstract

A preliminary investigation revealed that most consumers (72.6%) rated a given set of restaurants as either very low or low in terms of their sanitary quality. Additionally, it was discovered that even the most hygienic restaurants as rated by the eight hundred and eighty respondents had the highest morbidity rate (21.6%). This thus led to a detailed investigation into the extent to which a consumer's eating behavior was affected by the level of sanitation/morbidity rate of a restaurant in the set and the level of awareness of the eating population. The relationship between various sanitary indices, coliform counts and morbidity was also tested. The findings showed that the level of morbidity of the restaurants did not strongly affect patronage (consumer-eating behavior) (-0.49, $P < 0.01$). This highlights the need for greater public awareness. The sanitary indices strongly correlated with morbidity, with the appearance of food handler correlating the most (-0.93, $P < 0.01$). It is concluded that consumers may be at a high risk of infection. Therefore it is necessary to increase public awareness about the risk associated with eating in unhygienic restaurants.

1.0 Introduction

Food safety has always been a matter of global concern. As reported by the Food and Drug Administration (FDA), it is estimated that up to 76 million people get a food-borne illness each year in the United States alone. Additionally, 5000 people die from food-borne illness in the US annually with many cases of long-term morbidity (Willey *et al.*, 2008). In developing countries including Nigeria, the situation is even worse. Diarrhoea disease mostly caused by contaminated food and water is a major cause of morbidity and mortality in poor countries- particularly among children (Adams and Moss, 1995). A World Health Organization (WHO) Expert Committee report further states, 'In the contemporary world, food-borne illnesses are perhaps the most widespread problem and are an important cause of reduced economic productivity' (WHO, 2002).

The 1997 Local Government Health System profile for Nigeria on reported leading causes of mortality in different zones showed that diarrhoea cases accounted for 25% of mortality, followed by malaria 21%. The diarrhoea diseases may not be unconnected with food-borne disease (FAO/WHO, 2005). In Nigeria, a part of the problem is as the Secretary Of States, Hillary Clinton puts it, 'a lack of good governance' (The Guardian, Aug. 20, 2009). There have been different health policies formulated over the years stating the government's commitment to health for all. In a paper presented in the FAO/WHO Regional Conference on Food Safety 'The 1986 national health policy has been reviewed twice, first in 1988 and then in 1996.

In 1996, a national plan of action was drawn to cover the period 1996 to 2005, and this has now been adjusted in line with vision 2010' (FAO/WHO). Obviously, their money is never where their mouth is.

In the policy drafted, the local government authority is responsible for ensuring food safety for street food-vendoring, bukateria, catering establishments, and traditional markets. Unfortunately, there is a general lack of sanitation in these food establishments. On the part of scientific research conducted in this respect, seemingly more attention is given to coliform isolation rather than morbidity/epidermological investigations. This is a serious deficiency (Arduser and Brown,2005). It was against this backdrop a preliminary investigation was conducted on eight hundred and eighty respondents and ten restaurants, to ascertain among other things, the level of morbidity from these eating-places. The investigation revealed that most consumers (72.6%) rated a given set of restaurants as either very low or low in terms of their sanitary quality. Additionally, it was discovered that even the most hygienic restaurants as rated by the eight hundred and eighty respondents had the highest morbidity rate (21.6%). The population that reported cases of morbidity stood at 21.5%.

This thus led to a detailed investigation into the extent to which a consumer's eating behavior (patronage) was affected by the level of sanitation/morbidity rate of a restaurant in the set and the level of awareness of the eating population. The relationship between various sanitary indices, coliform counts and morbidity was also tested. Faecal coliform counts was used as an indicator of faecal contamination (WHO,2002). The findings of the study may be useful in developing strategies for improving Primary Health Care in developing countries and in generating better research methods that are people-oriented.

2.0 Materials and Methods.

2.1 Data Source

Respondents (200) who regularly eat from a given set of seven restaurants were asked to give their candid responses to six questions. As a result of the nature of the questions, the initial 880 respondents were reduced to 200, and the 10 restaurants to 7. This was to reduce the analytical problems associated with an overpopulated sample. All of the respondents were undergraduates who could correctly express themselves in writing, and the questions were kept simple. The restaurants selected were all located inside the university campus in Abraka, Delta State Nigeria. These restaurants regularly prepare and serve different local recipes to the university population. The participants could easily associate with the questions. They were required to answer the questions at the point of contact while giving them sufficient time and assistance, to minimize errors. It was from the restaurants that food samples were collected and analyzed for bacterial indicators of contamination. The restaurants were also physically investigated from the sanitary conditions of the kitchens to the personal hygiene of the food handler.

2.2 Measures

Interviews based on well-formulated questionnaire were conducted on one-on-one basis in lecture halls. There were six questions in total. The first question was on the extent to which the various restaurants were patronized. Likert-styled scale options (Very Often, 5; Often, 4; Slightly Often, 3; Rarely, 2; Never, 1) were provided for the participants. Afterwards the participants were requested to assess the eight restaurants on the basis of four sanitary indices. Thus the second question was on the assessment of the cleanness of the kitchen by the respondents. The Likert Scale ranged from 'Very Clean to Very Dirty' scoring (5-1) accordingly. The cleanness of the environment was similarly investigated with the scale ranging from 'Very Satisfactory to Very Poor'. The fourth question examined the appearance of food handlers following the Likert-styled Scale above. The last sanitary index assessed was the safety of water served in the restaurants. The scale ranged from 'Very Safe to Very Risky'. There is a maximum of 20 points using the four sanitary indices and a low score indicates very poor sanitation. Finally the frequency of morbidity (diarrhoea, and/or vomiting) from the respective restaurants was investigated. The Likert

scale scored (5-1) ranging from 'Very Often to Never' respectively. A high score indicated very poor sanitation.

2.3 Laboratory Analysis

Served water, as well as soup/stew samples from the restaurants under survey, were collected in sterile, screw-capped bottles (in triplicates) and transported immediately to the laboratory in ice-packs, for bacteriological (faecal coliform counts) analysis. The standard membrane filtration technique using 100mL of water samples, was adopted for detection as well as enumeration of faecal coliforms. Filters were layered on MacConkey agar (MA) after filtration, and incubated at 44.5°C for 24-48h as recommended by the WHO, (2002) and previously described by Ejechi and Ejechi, (2008). For stew/soup samples, 1mL of serially diluted samples was seeded onto the MA plates, well homogenized and incubated at 44.5°C as for water samples. In both cases, typical red/pink colonies were counted. Water samples having a single presence of faecal coliform, was regarded as contaminated (WHO, 2002). Faecal coliform counts per 100mL of soup/stew was extrapolated and similarly interpreted.

Randomly picked representative typical coliform colonies were selected for biochemical tests (IMViC) and Gram-staining procedure, in order to confirm their identity. The above procedure was repeated for the individual samples collected weekly from each restaurant, for 10 weeks.

2.4 Analysis of Data

Bar charts were used to show the variation in patronage and morbidity. A line graph was equally plotted to compare water quality and morbidity values. The sanitary indices and morbidity scores were analyzed by descriptive statistics while Duncan's Multiple Range Test (DMRT) was used in identifying the restaurants that were significantly different in values. Correlation test was performed with the sanitary indices, morbidity and patronage to ascertain the relationship between them. The mean values of the quantities were used for the correlation test.

3.0 Results and Discussion

Results show that morbidity did not significantly affect the frequency of visit by consumers to various restaurants (Figure.1). Rather, restaurants with relatively high morbidity counts had higher patronage. For instance, BR Restaurant had the highest patronage, which was consistent with the result from the preliminary investigation (38.9% patronage) but, did not have the lowest morbidity. It was rather at the middle of the profile. Conversely, MO with the highest morbidity had more patronage than PR- one of the restaurants with the lowest morbidity. While this was the general pattern observed, two restaurants reflected a different trend. The SG and ME restaurants had remarkably high patronage with comparably low morbidity counts. SG restaurant was the second most patronized and similarly, the second lowest cause of morbidity. The inference however seems to be that a consumer's decision to eat in any given restaurant is hardly considerably affected by the level of morbidity. Anyanwuocha (2006) states that price of a commodity, taste and fashion highly dictates the level of patronage. This may be true since consumers did not prioritize morbidity. This ignorance amongst consumers may account for the 21.6% morbidity rate initially discovered in the eating class.

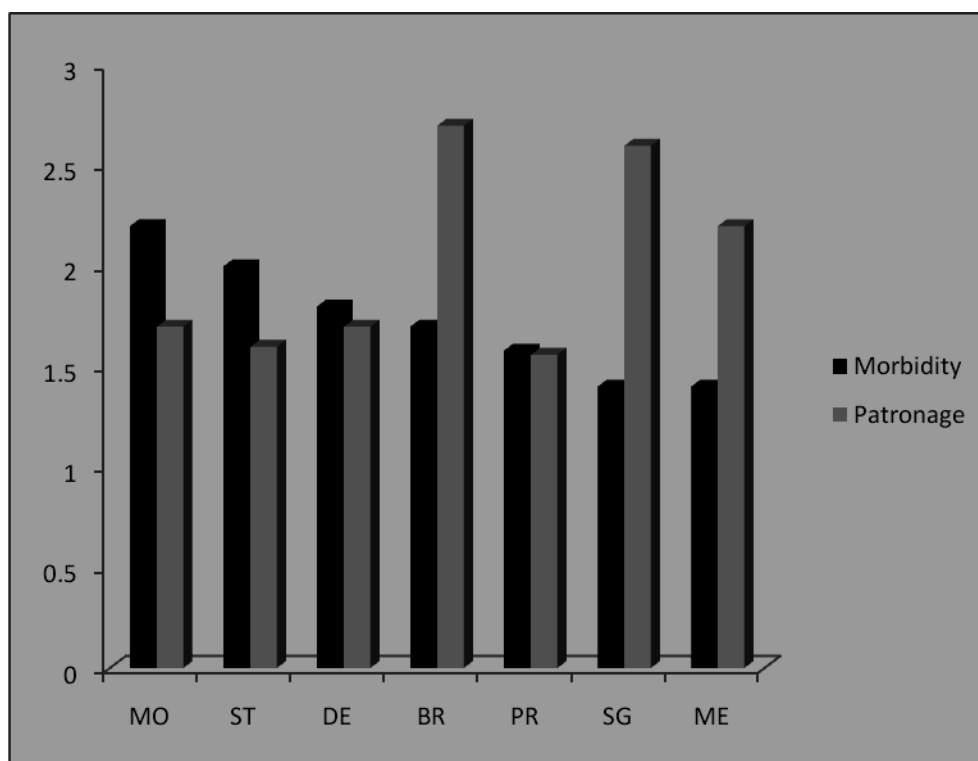


FIG 1: Comparison of Morbidity(series1) & Patronage(series2) Values of Restaurants

Figure 2 attempts to compare water safety levels with morbidity. The faecal coliform count was used to evaluate the safety of water, in an effort to uncover the cause of morbidity. Faecal coliforms are a subset of coliform bacteria commonly employed as indicators of faecal pollution in water and food. They inhabit the intestine of warm-blooded animals (Willey *et al* 2008). Generally, the coliform counts showed zero levels of faecal coliform per 100mL of water. WHO,(2002) states that 100ml of drinking water should contain no coliform. It is important to note though, that the water source serves the entire student/staff population of the campus, and no epidemic has been reported so far. However, a case of contamination was discovered in a single sample collected from ST restaurant. Besides this, it does seem safe to assume that morbidity did not result from water contamination. It became important to isolate restaurants that were significantly different in sanitary quality using the four sanitary indices. This served as a prelude to testing if this difference was portrayed in the level of morbidity and faecal coliform level of the food served.

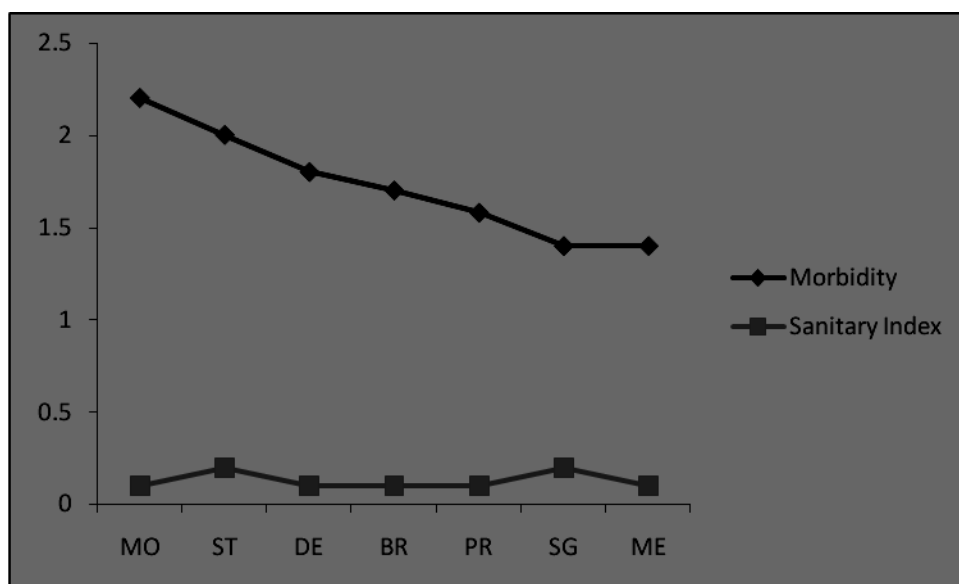


FIG2: Morbidity (MOR) and Sanitary (SAN) Index (faecal coliform in water) of the Restaurants

Table 1 shows that the significantly different restaurants through the table were MO and ST. They were significantly low in cleanness of kitchen and appearance of food handlers. Only ST restaurant was statistically different in cleanness of environment and even safety of water. The most clearly demarcated difference was in the appearance of food handler. This puts under question the personal hygiene of the handlers in both restaurants. Arduser and Brown (2005) states that personal hygiene is the best way to stop bacteria from contaminating and spreading into new areas. According to the CDC'S Surveillance for Food-borne Disease Outbreaks, (1988-1992), infected people touching food is responsible for 20% of food-borne outbreaks (CDC, 1999). ME and SUG spot were significantly high in all indices. This difference was more evident in the cleanness of kitchen in which they recorded significantly high counts with Great Delight restaurant. Arduser and Brown (2005) also states that keeping all food preparation areas clean and sanitized is an essential Standard Operating Procedure (SOP) to avoid food contamination. The sanitary assessment of the restaurants uncovered some probable causes of infection.

Furthermore, Table 2. shows that MO and ST restaurants were different in the total sanitary indices and in morbidity. The faecal coliform count to access food quality was a confirmation to this difference. There was evidently a problem with these two eating-homes. There was also a general problem in sanitation, revealed from physical evaluation of all the restaurants, although some restaurants looked better. The mean morbidity level was 1.76. Morbidity in this context meant that people either vomited or had diarrhoea after eating from these restaurants. This is a 19% rate of morbidity. The Centers of Disease Control and Prevention (CDC) have consistently stated that where reported, food-borne outbreaks were caused by mishandling of food, most of which occurred within the retail segment of the food industry where ready-to-eat food is prepared and provided to the public for consumption. (CDC, 2003)

All these food-borne diseases are associated with poor hygiene practices. Whether by water or food transmission, faecal-oral route is maintained, with the food providing the vital link between hosts (Willey *et al.*, 2008).

TABLE 1: SIGNIFICANTLY DIFFERENT RESTAURANTS IN SANITARY INDICES

Parameters	BR	PR	MO	ST	DE	ME	SUG
Cleanness of kitchen	2.97±0.79 ^{ac}	3.19±1.02 ^{acd}	2.82±1.04 ^{bc}	2.50±0.76 ^b	3.32±0.73 ^{ade}	3.67±0.90 ^{de}	3.55±0.79 ^{de}
Cleanness of environment	3.11±1.00 ^{ac}	3.04±1.04 ^{ac}	2.83±1.25 ^c	2.32±0.81 ^b	3.09±0.79 ^{ac}	3.73±1.07 ^d	3.53±0.90 ^{ad}
Appearance of handler	3.29±0.71 ^{ac}	3.08±1.12 ^a	2.53±0.98 ^b	2.63±0.79 ^b	3.34±0.76 ^{ac}	3.59±0.91 ^{ac}	3.5±10.86 ^{ac}
Safety of water	2.93±0.8 ^{ac}	3.12±1.07 ^{ac}	2.68±1.22 ^c	2.15±0.78 ^b	3.23±0.8 ^a	3.31±1.03 ^a	3.27±0.98 ^a

Means with the same asterisks are not significantly different (P=0.05) using Duncan's Multiple Range Test (DMRT).

TABLE 2: ASSESSMENT OF MORBIDITY, TOTAL SANITARY INDEX AND FOOD COLIFORM (FAECAL) LEVELS OF THE RESTAURANT

Parameters	BR	PR	MO	ST	DE	ME	SUG
Morbidity	1.74±0.99 ^{ab}	1.63±1.01 ^{ab}	2.18±1.49 ^b	2.00±1.17 ^b	1.85±1.20 ^{ad}	1.45±0.87 ^a	1.46±0.96 ^a
Tot San Index	12.30±2.08 ^a	12.20±2.71 ^a	10.68±2.90 ^b	9.65±1.98 ^b	12.80±1.62 ^a	14.41±2.19 ^c	13.82±1.91 ^{ac}
Coliform(Food)	1.50±0.45 ^a	1.46±0.30 ^a	1.80±0.50 ^b	1.77±0.46 ^b	1.60±0.53 ^{ac}	0.80±0.20 ^{ac}	1.00±0.21 ^{ac}

Means with the same asterisks are not significantly different (P=0.05) using Duncan's Multiple Range Test (DMRT).

Table 3 affirms the strong relationship between morbidity and most sanitary indices ($P < 0.05$). The strongest association is between morbidity and appearance of food handlers. The food handler is indeed very important in the spread of infection. Patronage correlated poorly with the sanitary indices and morbidity ($P > 0.05$). The implication is that while there is a proven relationship between sanitary quality and morbidity rate, consumers are generally not affected by this association in deciding which restaurant to patronize. Consumers clearly demonstrated a sound knowledge of the inadequate sanitary quality of the restaurants. 72.6% of the respondents in the preliminary exercise classified the set of restaurants as either very low or low in hygiene. The latter investigation affirmed that there was a 19% morbidity frequency. Yet consumers were generally not affected by this trend in their choice of patronage.

TABLE 3: ASSOCIATION BETWEEN SANITARY INDICES, MORBIDITY AND PATRONAGE

SANITARY INDICES	CORRELATION COEFFICIENT (r)	
	MORBIDITY	PATRONAGE
Cleanness of kitchen	-0.83	0.34
Cleanness of environment	-0.82	0.53
Appearance of food handler	-0.89	0.58
Safety of Water	-0.74	0.35
	0.92	0.49
Faecal count (food)	0.39	0.35
Faecal count (water)	-	-0.49
Morbidity		

The root problem may be distrust in the authority. Some respondents reacted as follows, when asked to fill the questionnaire:

‘ Will this research change anything? There should be a monitoring team but who will provide that?’

Under the Constitution of the Federal Republic of Nigeria, 1999, Section 7; the local government is responsible for the control and regulation of the restaurants and other places of sale of food to the public (Afrobarometer, 2008). Afrobarometer further states: “ Over the decade, under the elected civilian administration, local government councils in Nigeria have received substantial revenue allocations without always providing commensurate services.” Indeed as Jeff Jarvis, 2009 states: “ the crisis the world is suffering is a failure in leadership (Davos Diary, 2009). The Nigerian public clearly considers that the local government has failed in the performance of their primary responsibilities, which includes service delivery. The above publication in a survey conducted states that 72% of the Nigerian public expressed distrust in the local government. Kathryn Whetten *et al.*, (2009) states that distrust may be a barrier to optimal health. Since consumers must eat they rather put up with the morbidity than starve. Therefore it is the respondents’ dissatisfaction with the government that has led to an adaptation to the malfunctioning system.

4.0 Conclusion

The 'clashes and flashes' of the Niger-delta manifests, not only in youth militancy as a sequel to oil exploration and exploitation, but also in heightened commercial activities, road-side restaurants, not an exception. Considering our findings that individuals are to a large extent aware of the deficiencies in sanitary conditions of most restaurants yet, patronage was not affected by cases of morbidity, there are at least two fundamental problems. First, is the fact that individuals could readily sacrifice threats to their health, on the alter of monetary conservation and hence, needs to be rescued from themselves. Second, is the painful realization that the agent of Government saddled with the responsibility of monitoring and regulating food safety of restaurants, have failed.

This therefore, serves as a wakeup call, both to the regulatory agencies in particular, and individuals in general, that we must not wait for an epidemic, to rise up to the challenge. There is an urgent need for individuals to translate their knowledge of poor sanitary indices, to proper judgment of patronage that is not predicated solely on cost of the food served and sold. Furthermore, it is high time the Local Government activated that arm of herself charged with the control and regulation of restaurants, and other places of food sale to the public. A stitch in time, saves nine.

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ANTIMICROBIAL EFFECT OF PHYLLANTHUS AMARUS AND SYNTHETIC DRUGS ON SELECTED CLINICAL ISOLATES: A COMPARATIVE EFFICACY

***¹Ojo S.K.S, ²Alikwe P.C.N., ¹Awokoya O.O. and ¹Okowa T.B.**

^{*1} Dept. of Biological Sciences, Novena University, Ogume, Delta State.

² Dept. of Chemical Sciences, Novena University, Ogume, Delta State.

*Corresponding author: OJO S.K.S E-mail: gloriousstephen@yahoo.com

Abstract

Herbal medicines tend to look primitive and unscientific when compared to synthetic (conventional) drugs, which are thought to be more reliable than those made from plants. The aim of the study is to compare the effectiveness of the two antimicrobial agents vis-à-vis the solvent of extraction. The antimicrobial activity of *Phyllanthus amarus* with commercial antibiotics were tested against 4 bacterial strains (*Staphylococcus aureus*, *E. coli*, *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*) and 1 fungal strain (*Candida albicans*). The absolute ethanol extracts were obtained by Soxhlet extraction and the local (gin) ethanolic extracts were obtained by cold percolation method. The results indicated that the plant extracts exhibited antimicrobial activity against one or more of the tested microorganisms at the concentrations of 200, 100 and 50mg/ml. Absolute ethanolic extracts demonstrated the highest inhibitory effect and none of the extracts had inhibitory effect against fungal strain. Of the standard antibiotics tested, Ciprofloxacin and Streptomycin demonstrated the highest inhibitory effect against Gram negative and Gram positive bacteria respectively. Nystatin (antifungal agent) and the plant extracts had no inhibitory effect against *Candida albicans*. This study showed that *Phyllanthus amarus* extracts are best global alternative therapy in the face of drug resistance challenges and the perturbing drug side-effects. Therefore, herbal potency should be harnessed into developing new standard drugs.

Key words: *Phyllanthus amarus*, synthetic drugs, antimicrobial activity, clinical isolates

INTRODUCTION

Therapeutic values of natural products (medicinal plants) has been an increasing interest world-wide with the belief that the cure to any debilitating human ailments and diseases may be found among the world's flora in nature's pharmacy (Olowosulu and Ibrahim, 2006). There are multitudes of potential useful bioactive substances to be derived from these plants and these phytochemical have made significant contribution in maintaining human health (Oluwafemi and Debiri, 2008; Duraipandiyan *et al.*, 2006). The significance of drugs derived from plants cannot be over-emphasized with the recent trend of high percentage of resistance of microorganism to the present day antibiotics (Ibekwe *et al.*, 2000). Sustainable management of traditional medicinal plant resources is important, not only because of their value as a potential source of new drugs but due to reliance on traditional medicinal plants for health. The vast majority (70-80%) of people in Africa consult traditional medical practitioners (TMPs) for health care. With few exceptions, traditional medicinal plants are gathered from the wild. However, increasing demand for popular herbal medicines is expected in the foreseeable future (Cunningham, 1993).

Phyllanthus amarus (Schumacher and Thonn) is a herb belonging to the family Euphorbiaceae, consisting of 300 genera and 6500 species which is commonly used in Central and Southern India and are also found in other countries including China, Philippines, Cuba, Nigeria and Guam (Oluwafemi and Debiri, 2008). It

grows up to 15-80 cm in height, all parts of the plant are employed therapeutically. It is used in the treatment of malaria-related symptoms, jaundice, constipation, diarrhea, diabetes, flu, kidney ailments, chronic dysentery, frequent menstruation, ringworm, ulcer, genito-urinary infection, haemorrhoids and gonorrhoea (Hanumanthacar *et al.*, 2007; Lim and Murtijaya, 2007). *P. amarus* extracts were found to possess anti-viral property against hepatitis B virus (HBV), inhibit DNA polymerase activity of HBV and suppress its mRNA transcription, translation and its replication. It also inhibits HIV-1 replication in cell culture, HIV reverse transcriptase, and block virus uptake. It also blocks HIV-1 attachment to its primary cellular receptor CD₄ and the HIV-1 enzymes integrase, reverse transcriptase and protease (Lim and Murtijaya, 2007). *P. amarus* is also used to treat hepatic and urolitic diseases, possess diuretic activity, antitumour, anticancer, hepato-protective, antioxidant and anti-inflammatory activity. It mainly contains phyllanthin and hypophyllanthin as active ingredients and the aqueous extract has been employed for treatment of nervous debility, epilepsy and used as medhya (intellect promoting) (Hanumanthacar and Milind, 2007; Lim and Murtijaya, 2007). This study tends to compare the effectiveness of *P. amarus* and synthetic drugs on the selected clinical isolates vis-à-vis the solvent of extraction.

MATERIALS AND METHODS

Source of *Phyllanthus amarus*

Fresh plant materials were collected from Novena University, Ogume (Amai Campus), Delta State, Nigeria. The botanical identity was determined and authenticated at the Department of Biological Sciences, Novena University, Ogume.

Processing/Extraction of Plant Sample

The whole plant was harvested, rinsed properly with tap water and sterile distilled water. The various parts of the plant were air-dried, pounded separately with a mortar and pestle and micronized with the aid of a blender to powdered form. 25g of the pulverized plant was extracted in 250ml of absolute (purified) ethanol using a Soxhlet extractor for 6-8h and 15g of the pulverized powdered plant was extracted in 200ml of local (gin) ethanol at room temperature for 48h (cold percolation). A sieve was used to filter the plant residues and the filtrate obtained was further purified by filtering through Whatman No.1 filter paper (Atata *et al.*, 2003). The stock solution of extract was sterilized by filtration using Millipore filter membrane (pore size 0.45µm diameter) (Ronald, 1995). The sterile extracts were stored at 4°C in a refrigerator till required for use. Various concentrations of plant extracts were obtained which are: 200, 100 and 50mg/ml.

Sterility Proofing of the Extracts

The extracts were checked for sterility after Millipore membrane filtration by introducing 2ml of this supposed sterile extract into 10ml of sterile Mueller-Hinton broth. Incubation was done at 37°C for 24h. A sterile extract was indicated by absence of turbidity or clearness of the broth after the incubation period (Ronald, 1995).

Test Organisms

The test organisms used for this study were *Candida albicans* (fungi), *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Escherichia coli*. The pure clinical isolates were obtained from the Medical Microbiology department, University of Benin Teaching Hospital, Benin, Edo State. All clinical isolates were checked for purity and maintained on Nutrient agar (for fungi on PDA) slant at 4°C in a refrigerator till required for use.

Standardization of Test Organisms

Two colonies each of the test organisms were inoculated into sterile test tubes containing Mueller-Hinton broth and incubated at 37°C for 24h. The turbidity indicated by this organism was adjusted to match the turbidity (opacity) standard as described by Cheesbrough (2004).

Antimicrobial Sensitivity Testing

This was carried out using the agar well diffusion method and disk diffusion method as described by Hugo and Russel (1996). The plant extracts and the multiple antibiotic disks (for Gram positive and negative organisms) were used on separate plates with extracting solvent as control. The experiment was performed in triplicates and the average readings were taken as the zone of inhibition.

Determination of Minimum Inhibitory Concentration (MIC)

The MIC of the extracts against the test organisms were determined by using the Broth dilution method. Ten test tubes containing 9ml each of sterile Mueller-Hinton broth was prepared. 1ml of the working extract (200mg/ml) was transferred into the first test tube of 9ml nutrient broth and serial dilution was carried out to the 7th test tube to obtain different concentrations (200mg/ml, 100mg/ml, 50mg/ml) (Ibekwe *et al.*, 2001). Each dilution was inoculated with 0.1ml of the standard bacterial and fungal cell suspension separately and incubation was done at 37°C for 24h. The growth of the inoculum is indicated by turbidity or cloudiness and the lowest concentration of the extract which inhibited the growth of the test organism was taken as the MIC. Negative controls were set up as follows: test tube 8- Mueller-Hinton broth only; 9th test tube- Mueller-Hinton broth and sterile plant extract and a positive control containing Mueller-Hinton broth and a test organism (Sule and Agbabiaka, 2008).

Determination of Minimum Bactericidal and Fungicidal Concentration (MBC and MFC)

The MBC and MFC were determined by taking a loopful from each test tube showing no growth during MIC determination and plated on Mueller-Hinton agar, incubated for 24h at 37°C. The least concentration showing no growth was noted as the MBC and MFC (Mbata and Saikia, 2008).

Statistical Analysis

Data was analysed using one sample T-test. Results expressed as mean \pm standard deviation. All analysis were done using SPSS version 16.0.

RESULTS

The antimicrobial activity of *P. amarus* in this study showed the zones of growth inhibition of absolute ethanolic extracts (leaf, stems and roots) on the test organisms (Table1). On the contrary, the local (gin) ethanolic extracts of *P. amarus* leaves showed no zones of growth inhibition on test organisms except *Pseudomonas aeruginosa* (Table 2). The results of the MIC for absolute ethanolic leaf extract showed that majority of the test organisms were inhibited at 100mg/ml with the corresponding diameter for zones of inhibition between 19.0-21.8mm (Table 3&1). The MIC for absolute ethanolic stem and root extract also showed inhibition at 50mg/ml for majority of the test organisms and the zones of growth inhibition between 19.0-21.5mm (Table 3&1). The MIC for local (gin) ethanolic leaf extracts indicated at 50mg/ml had zones of growth inhibition between 19.0-20.0mm for *Pseudomonas aeruginosa* (Table 4&2). Table 5 showed the activity of standard (commercial) antibiotics on test organisms with ciprofloxacin having the highest activity for the Gram negatives; Streptomycin for the Gram positive and no activity seen in Nystatin for *Candida albicans*.

Table1. Zones of growth inhibition (mm) of absolute ethanolic extracts of leaves, stems and roots of *P. amarus* on tested organisms

Test organisms	Absolute leaf extract (mg/ml)			Stem & root extract (mg/ml)		
	200	100	50	200	100	50
<i>E. coli</i>	20.5±0.05	19.0±0.10	NI	21.5±0.13	20.5±0.5	19.0±0.0
<i>S. aureus</i>	22.5±0.05	21.5±0.13	NI	22.7±0.51	21.7±0.14	20.0±0.1
<i>K. pneumoniae</i>	22.8±0.13	21.8±0.12	21.0±0.0	23.0±0.1	22.0±0.1	21.5±0.13
<i>P. aeruginosa</i>	NI	NI	NI	20.0±0.1	19.0±0.0	NI
<i>Candida albicans</i>	NI	NI	NI	NI	NI	NI

Key: NI- No inhibition at the concentration used.

Table2. Zones of growth inhibition (mm) of local (gin) ethanolic extracts of leaves, stems and roots of *P. amarus* on tested organisms

Test organisms	Absolute leaf extract (mg/ml)			Stem & root extract (mg/ml)		
	200	100	50	200	100	50
<i>E. coli</i>	NI	NI	NI	NI	NI	NI
<i>S. aureus</i>	NI	NI	NI		NI	NI
<i>K. pneumoniae</i>	NI	NI	NI	21.0±0.89	20.2±0.1	19.0±0.0
<i>P. aeruginosa</i>	20.05±0.5	20.0±0.1	19.0±0.1	21.0±0.89	20.5±0.5	20.0± 0.1
<i>Candida albicans</i>	NI	NI	NI	NI	NI	NI

Key: NI- No inhibition at the concentration used.

Table3. Determination of Minimum Inhibitory Concentration (MIC) of the absolute ethanolic leaf, stem & root extracts of *P. amarus* on tested organisms.

Test organisms	Concentration (mg/ml)					
	Absolute leaf extracts			Absolute stem & root extracts		
	200	100	50	200	100	50
<i>S. aureus</i>	+	+	-	+	+	+
<i>E. coli</i>	+	+	-	+	+	+
<i>K. pneumoniae</i>	+	+		+	+	
<i>P. aeruginosa</i>	-	-	-	+	+	-
<i>Candida albicans</i>	-	-	-	-	-	-

Key: + : Inhibition, - : No inhibition

Table4. Determination of Minimum Inhibitory Concentration (MIC) of Local (gin) ethanolic leaf, stem & root extracts of *P. amarus* on tested organisms.

Test organisms	Concentration (mg/ml)					
	Local (gin) leaf extracts			Local(gin) stem & root extracts		
	200	100	50	200	100	50
<i>S. aureus</i>	-	-	-	-	-	-
<i>E. coli</i>	-	-	-	-	-	-
<i>K. pneumoniae</i>	-	-		+	+	
<i>P. aeruginosa</i>	+	+	+	+	+	+
<i>Candida albicans</i>	-	-	-	-	-	-

Key: + : Inhibition, - : No inhibition

Table5. Zones of growth inhibition of standard antimicrobial agents on test organisms.

Standard agents	<i>K. pneumoniae</i>	<i>P. aeruginosa</i>	<i>E. coli</i>	<i>S. aureus</i>	<i>C. albicans</i>
Septin	NI		NI	12mm	ND
Chloramphenicol	NI		NI	17mm	ND
Sparfloxacin	NI		NI	NI	NI
Ciprofloxacin	22mm		18mm	13mm	ND
Amoxacillin	NI		NI	NI	ND
Augmentin	NI		NI	NI	ND
Gentamicin	NI		NI	NI	13mm
Perfloxacin	18mm		6mm	NI	ND
Streptomycin	1mm		NI	12mm	22mm
Tetracycline	ND		ND	ND	8mm
Ampicillin	ND		ND	ND	NI
Penicillin	ND		ND	ND	NI
Erythromycin	ND		ND	ND	20mm
Cloxacillin	ND		ND	ND	NI
Nystatin	ND		ND	ND	NI

Key: NI – No inhibition; ND – Not determined

DISCUSSION

Several studies had reported that plants contain antimicrobial substances (Oluwafemi and Debiri, 2008; Sule and Agbabiaka, 2008; Akujobi *et al.*, 2004 and Ibekwe *et al.*, 2001). The results of this present study confirm the reports of these previous workers. The result showed that local (gin) ethanolic extracts of *Phyllanthus amarus* is not as effective as the absolute ethanolic extracts.

Though, it is believed that ethanol is generally able to dissolve multivariable types of compounds: polar and non-polar, simple and complex chemical structures compared with chloroform which solubilizes mainly flavanols - Phenolic compounds from plant (Cowman, 1999). Hence, the low result in local gin (usually used by local populace) could be as a result of impurities affecting the solubility of the plants phytochemicals during extraction. Thus, the relative amount of phytochemical substances from plant extraction depends on the solubility of the phytochemical in the solvent used in extraction (Olowosulu and Ibrahim, 2006).

Although fifteen commercial (standard) antibiotics were employed against the test organisms, only Ciprofloxacin, Perfloxacin, Chloramphenicol, Streptomycin and Erythromycin had antimicrobial effect in decreasing order. Ciprofloxacin had the highest zone of growth inhibition against *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* respectively. Remarkably, ciprofloxacin has been the antibiotic of choice for the treatment of Gram negative bacterial infections but *E. coli* was resistant to it in this study, which could either be the strains of *E. coli* used or self-medication as practiced by patients. This result is in conformity with an earlier report by Oluwafemi and Debiri (2008). Streptomycin had the highest zone of growth inhibition against *S. aureus* as compared to other antibiotics. These possibly could be as a result of methicillin resistant strains of *S. aureus* (MRSA), which is associated with skin and respiratory diseases. This study is in consonance with Akinyemi *et al.* (2005), which showed that *P. amarus* (variant of *P. discoides*) was active against *S. aureus* and MRSA. It could be deduced from this study that *P. amarus* is more effective as compared to the commercial antibiotics. It is also evident from this study that *P. amarus* cannot be regarded as an anticandidal agent.

CONCLUSION

Our investigation on *Phyllanthus amarus* as an antimicrobial agent in Nigeria ethno medicine only justifies its use against bacterial infection and not candidal infection. Also, the traditional use of local gin as an extracting solvent does not permit the full extraction of the active ingredients of the plant as compared with the absolute ethanolic extraction. Therefore, in vivo study on this medicinal plant is necessary as to determine toxicity of the

active constituents, their side effects, serum-attainable levels, pharmacokinetic properties and diffusion in different body sites. Thus, *P. amarus* is an effective and alternative antimicrobial agent as compared to the synthetic drugs.

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RESPONSE OF SIX CULTIVARS OF COWPEA (*Vigna unguiculata* (L.) WALP) TO SPENT ENGINE OIL

Agbogidi O. M.

Department of Forestry and Wildlife, Faculty of Agriculture, Delta State University, Asaba Campus
omagbogidi@yahoo.com; Phone: +2347038679939; +2348056306219

Abstract

Laboratory studies were carried out in 2008 at the Plant and Soil laboratory of the Faculty of Agriculture, Delta State University, Asaba Campus to investigate the response of six cultivars of cowpea (IT80D-699, IT82 (e-18), IT84S-2246-4, TVx3236, IT90K-277-2 and IT870-94-1) to spent engine oil. Seeds of the six cultivars of cowpea were presoaked in water for 24 hours and thereafter, soaked in spent engine oil for varying hours (0, 1, 2, 4, 8 and 16). Other seeds were soaked in the oil for 0, 1, 2, 3 and 4 days before germinated in Petri dishes lined with moist tissue paper. The results showed that the percentage germination, days to germination and rate of germination of the cowpea cultivars were both periods of seed soaking in oil and cultivar dependent. The results also showed that the longer the hours of seed presoaking in the oil, the poorer the germination response of the seeds. No germination occurred while seeds were still in the oil. TVx3236 followed by IT84S-2246-4 were more tolerant among the cowpea cultivars examined hence could be considered for phytoremediation practice in oil producing areas of the Niger Delta.

Keywords: *Vigna unguiculata*, germination response, spent engine oil

Introduction

Cowpea is an annual, herbaceous legume. It is a short-day crop sensitive to chilling temperatures but adapted to warm weather and humid conditions (Islam *et al.*, 2006). It belongs to the family Fabaceae and sub-family Faboidea. The Yoruba in Nigeria locally calls it “wake” by the Hausa tribe and “ewa”. It originated from central Africa but it is now widely cultivated in many parts of the tropics and sub-tropics including west Africa and India (Olafeke *et al.*, 2006). The leaves of cowpea are eaten in salad and the immature pods are used as vegetable. The grains are rich source of plant protein to man; they contain mineral salts, vitamins and fats. The young shoots are eaten like spinach (Adepoju *et al.*, 2006) while the immature seeds are eaten fresh, frozen and canned. It is regarded as poor man’s meat because they are the cheapest source of protein; essential amino acids in them are also in sufficient amount (Awe, 2008; Omotugba *et al.*, 2008). Their high nutritional value characteristics make cowpea a candidate for controlled ecological life support system (CELSS), which uses green plants to supply food, oxygen and purified water for inhabitants of future space, craft and planetary bases. The crop has other uses such as in livestock feed and enhancement of soil fertility and as a cover crop in agriculture. Because of its nutritional value to man and livestock, the cultivation of cowpea in recent times has increased tremendously.

Various petroleum products are common soil contaminants and often contain potentially hazardous chemicals especially the polycyclic aromatic hydrocarbons (Sharifi *et al.*, 2007). Spent engine oil, usually obtained after servicing and subsequently draining used oil from automobiles and generator engines, are indiscriminately disposed into gutters, water drains, open vacant plots and farms in Nigeria by auto-mechanics and allied artisans with workshops on the road sides and open places (Anoliefo and Vwioko, 2001). Atuanya (1987) and Agbogidi and Ejemete (2005) noted that oil in soil has deleterious effects on

the biological, chemical and physical properties of the soil depending on the dose, type of the oil and other factors. Benka-Coker and Ekundayo (1995) and Benka-Coker and Ekundayo (1997) also reported that the microbiological components of soil are usually negatively affected when oil is applied to soil. Although, some research works have been conducted on the effects of spent lubricating oil on the germination of some economic crop plants (Anoliefo and Vwioko, 1995; Anoliefo and Vwioko, 2001; Anoliefo and Edegbai, 2000; Agbogidi and Nweke, 2006; Agbogidi *et al.*, 2006a; Agbogidi *et al.*, 2006b; Sharifi *et al.*, 2007), there is paucity of information on the response of cowpea to spent oil. The aim of this study was to evaluate the germination of six cowpea cultivars as affected by spent engine oil.

Materials and Methods

The study was carried out in 2008 at latitude $6^{\circ}14'N$ and longitude $6^{\circ}49'E$ at the Plant and Soil laboratory of the Faculty of Agriculture, Delta State University, Asaba Campus, Nigeria (Asaba Meteorological Office, 2008). The six cultivars of cowpea (IT81D-699, IT82 (e-18), IT84S-2246-4, TVx3236, IT90K-277-2 and IT870-94-1) were purchased as a single batch from the International Institute for Tropical Agriculture (IITA), Ibadan (Onne station) Oyo State, Nigeria while the spent engine oil was obtained as a pooled used engine oil from 12 different motor mechanic workshops in Asaba, Delta State. The seeds were subjected to viability test using floatation technique. Using randomized samples taken from homogenous population of each seed type, the six cultivars of cowpea were presoaked in water for 24 hours and thereafter, in (100%) spent engine oil for varying periods of time (0, 1, 2, 4, 8 and 16 hours) and then, germinated on moist tissue paper placed in Petri-dishes. The significant of seed presoaking in 100% SEO is that it closely equates the amount released to the environment in the event of disposals. In the same vein, the seeds of the six cultivars of cowpea were presoaked in sent engine oil (SEO) for 0, 1 2, 3, and 4 days and thereafter, germinated in Petri-dishes lined with moist toilet papers. Seeds soaked in the oil were removed at intervals and planted out in Petri dishes. Seed soaking was done in a large plastic bowl while the Petri dishes served as germinators. For each treatment, a total of 100 seeds was used and replicated four times. The set-up was arranged in a randomized complete block design. Readings were taken at 24 hours intervals following the procedure of Anoliefo and Vwioko (1995) and unequivocal emergence of ridicules was used as a critical stage of germination. Parameters determined were percentage germination, days to germination and rate of germination. Percent (%) germination was calculated using this formula. Number of seedlings that sprouted over/Number of seeds planted multiplied by 100 over 1. Days to germination were calculated by counting from the day of sowing to the day of unequivocal emergence of ridicules while the rate of germination was calculated based on when about 50% of the seeds planted germinated. Data collected were subjected to analysis of variance while the significant treatment means were separated with the Duncan's multiple range tests (DMRT) using SAS (1996).

Results and Discussion

All the seeds of the cowpea cultivars sown in the uncontaminated Petri-dishes germinated on the 5th day after sowing. Seeds sown in dishes after presoaking in water differed significantly ($P \leq 0.05$) in their germination records (Table 1). A significant ($P \leq 0.05$) reduction was observed in dishes with seeds presoaked in spend engine oil for varying hours. The reduction was observed to exposure-time dependence. Similarly, percentage germination of cowpea seeds from all the cultivars sown in the dishes without presoaking in SEO differed significantly at the 5% probability level (Table 1). The rate of germination of all the cowpea cultivars also shown a significant reduction ($P \leq 0.05$) as the period (hours) of seed presoak increased. The results have shown that oil has an acute effect on seed germination. The seeds soaked in the oil for 16 hours failed to germinate (Table 1).

The results also showed depression in the germination characteristics of the cowpea cultivars tested. No germination was recorded in cowpea seeds soaked in oil for more than 2 hours (Table 2). No

seeds germinated while still soaked in the oil. The oil could have endangered the life of the seed embryo and hence lead to loss of seed viability. This finding is in agreement with prior reports of Agbogidi and Nweke (2005), Siddiqui and Adams (2002) and Sharifi *et al.*, 2007.

The results also showed that TVx3236 and IT84S-2246-4 were more tolerant to the oil levels used in this study. Agbogidi and Nweke (2005), Agbogidi and Nweke (2006) had reported that oil effects on plants are species dependent. The herbicidal properties of oil on plants have also been reported by Adams and Duncan (2002), Agbogidi and Ofuoku (2005), Nwadinigwe and Uzodimma (2005) and Agbogidi and Eshegbeyi (2006).

Conclusively, the current study has demonstrated that spent engine oil has a significant effect of reducing the germination characteristics of the six cowpea cultivars tested with the TVx3236 and IT84S-2246-4 showing some levels of tolerance. The different sensitivity of plants to spent oil toxicity can be exploited in phytoremediation practice by choosing species that are well tolerant to the contaminant. TVx3236 and IT84S-2246-4 from the current study could be considered for phytoremediation of spent engine oil polluted sites especially at low concentrations. Such studies should be on increased accumulation (over soaking time) of oil metabolites in these cultivars tissues as well as the measurements of plant uptake and /or degradation of absorbed oil.

Table 1. Germination characters of six cultivars of cowpea as influenced by spent engine oil

	Period of seed presoaking in oil (Hours)						
Cowpea cultivars	0	1	2	4	8	16	Mean
% Germination							
IT81D-699	100.0	62.6	42.4	38.4	32.5	0.0	45.9d
IT82 (e-18)	100.0	67.4	48.7	40.2	36.4	0.0	48.8c
IT84S-2246-4	100.0	73.4	60.2	54.3	48.4	5.4	56.9b
TVx3236	100.0	78.1	69.0	58.4	50.6	15.7	61.9a
IT90K-277-2	100.0	57.6	40.2	37.8	30.7	0.0	44.4e
IT870-941-1	100.0	52.3	35.9	30.6	24.3	0.0	40.5f
Means	100.0a	65.2b	49.4c	43.3d	37.2e	3.5f	
Days to germination							
IT81D-699	5.7	6.6	6.9	7.3	7.6	0.0	5.7d
IT82 (e-18)	5.6	6.7	7.3	7.5	7.7	0.0	5.6c
IT84S-2246-4	5.0	5.5	6.7	6.9	7.1	7.3	6.4a
TVx3236	5.0	5.3	5.4	5.9	6.2	6.4	5.8c
IT90K-277-2	5.2	6.8	7.2	7.4	7.6	0.0	5.7d
IT870-941-1	5.5	6.9	7.4	7.7	7.9	0.0	5.9b
Means	4.5e	6.3d	6.3d	7.1b	7.4a	2.3f	
Rate to germination							
IT81D-699	10.0	7.4	6.3	5.2	5.0	0.00	5.7d
IT82 (e-18)	10.0	7.6	6.5	6.0	5.1	0.0	5.9c
IT84S-2246-4	10.0	8.6	8.2	7.9	7.4	6.2	8.1b
TVx3236	10.0	9.7	9.3	9.0	8.8	6.4	8.9a
IT90K-277-2	10.0	7.3	6.2	5.3	5.0	0.0	5.6d
IT870-941-1	10.0	7.4	6.1	5.1	4.8	0.0	5.6d
Means	10.0a	8.0b	7.1c	6.4d	6.0e		

Means with different letters of a parameter are significantly different at ($P \leq 0.05$) using DMRT.

Table 2. Germination records of the six cultivars of cowpea as affected by period of seed presoaking in SEO

	Period of seed soaking (days) in SEO before germination					
Cowpea cultivars	0	1	2	4	8	Mean
Percent germination						
IT81D-699	100.0	0.0	0.0	0.0	0.0	20.0c
IT82 (e-18)	100.0	0.0	0.0	0.0	0.0	20.0c
IT84S-2246-4	100.0	60.2	5.1	0.0	0.0	33.1b
TVx3236	100.0	70.2	10.7	0.0	0.0	36.1a
IT90K-277-2	100.0	0.0	0.0	0.0	0.0	20.0c
IT870-941-1	100.0	0.0	0.0	0.0	0.0	20.0c
Means	100.0a	21.7b	2.6c	0.0d	0.0d	
Days to germination						
IT81D-699	5.0	0.0	0.0	0.0	0.0	1.0c
IT82 (e-18)	5.0	0.0	0.0	0.0	0.0	1.0c
IT84S-2246-4	5.0	6.9	8.2	0.0	0.0	4.0a
TVx3236	5.0	6.0	6.7	0.0	0.0	3.5b
IT90K-277-2	5.0	0.0	0.0	0.0	0.0	1.0c
IT870-941-1	5.0	0.0	0.0	0.0	0.0	1.0c
Means	5.0a	2.2c	2.5b	0.0d	0.0d	
Rate to germination						
IT81D-699	10.0	0.0	0.0	0.0	0.0	2.0c
IT82 (e-18)	10.0	0.0	0.0	0.0	0.0	2.0c
IT84S-2246-4	10.0	7.1	3.0	0.0	0.0	4.0b
TVx3236	10.0	8.6	5.4	0.0	0.0	4.8a
IT90K-277-2	10.0	0.0	0.0	0.0	0.0	2.0c
IT870-941-1	10.0	0.0	0.0	0.0	0.0	2.0c
Means	10.0a	2.6b	1.4c	0.0d	0.0d	

Means in same column of a parameter with different letters are significantly different at ($P \leq 0.05$) using DMRT.

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ANTIMICROBIAL SUSCEPTIBILITY AND PLASMID PROFILES OF *PSEUDOMONAS AERUGINOSA* AND *STAPHYLOCOCCUS AUREUS* ISOLATED FROM BURN WOUND

S. O. Egbule*, B. Igere' and E. Enameguonor*

*Department of Microbiology, Delta State University, Abraka

' Department of Biotechnology, Western Niger Delta University, Oghara.

Abstract

Eighteen strains of *Pseudomonas aeruginosa* (16) and *Staphylococcus aureus* (2) isolated from hospitalized burn wound patients were selected for antimicrobial susceptibility and plasmid analysis. All *Pseudomonas aeruginosa*, were resistant to Septrin, gentamycin, amoxicillin, streptomycin, chloramphenicol and augmentin. All *Staphylococcus aureus* were resistant to septrin, perfloxacin, gentamycin, amoxicillin, ciprofloxacin, erythromycin, ampiclox, cefuroxime and ceftriazone. All strains were multidrug resistant (MDR). Sixty-one percent (61.1%) MDR isolates carried their resistance on plasmid. Plasmid profile analysis showed similar plasmid size of $\geq 1200\text{bp}$. The acquisition of similar plasmid may suggest phylogenetic relatedness.

Introduction

The increasing morbidity and mortality due to burn injury resulting from multidrug resistant (MDR) *Pseudomonas aeruginosa* and *Staphylococcus aureus* has been recognised in hospitalized patients. *Pseudomonas aeruginosa* is an important opportunistic pathogen with innate resistant to many antibiotics. Despite innate resistance, additional acquired resistance due to plasmids is also found. *Staphylococcus aureus* rapidly acquire resistance to different classes of antibiotics (Sleigh and Timbury, 1986). Strains of *Pseudomonas aeruginosa* and *Staphylococcus aureus* resistant to different classes of antimicrobial agent are endemic already in numerous hospitals and chronic burn unit institutions (Alice *et al.*, 1999; Sleigh and Timbury, 1986). Therefore acquired resistance to antimicrobial agents constitutes a major challenge for antibiotic therapy, especially when it is associated with resistance to many classes of antibiotics such as beta lactams, aminoglycosides and fluoroquinolones (Laura *et al.*, 2004; Livermore, 2002).

The association of *Pseudomonas aeruginosa* and *Staphylococcus aureus* in hospitalized burn wound worldwide has increased the importance of epidemiological studies. A number of plasmid DNA detection procedures have been used to analyse plasmid (Karunasagar *et al.*, 1987; Birnboim and Doly, 1979; Fujita *et al.*, 1994). This study was designed to investigate the occurrence of plasmid DNA carrying antibiotic resistance genes in *Pseudomonas* and *Staphylococcus*.

Materials and Methods

Collection of Samples

A total of 50 swabs were obtained from hospitalized burn wounds patients from General hospital, Warri. Ethical clearance and consent from the patients were got before sample collection. The swab sticks were transported in ice to Lahor laboratory for processing.

Bacteriology

The burn wound obtained were inoculated into MacConkey agar, blood agar and nutrient agar respectively and incubated at 37°C for 24 hrs. Bacterial colonies were later gram stained and characterized using standard bacteriological methods according to Cowan and Steel (1993).

Antibiotic Susceptibility Testing

Susceptibility testing was determined by agar disk diffusion methods as recommended by National Committee for Clinical Laboratory Standards (NCCLS) using Mueller Hinton agar. The disk used contained the following antibiotics: amoxicillin (30µg), gentamycin (10µg), pefloxacin (10µg), streptomycin (30µg), spetrin (30µg), ciprofloxacin (10µg), erythromycin (10µg), ampiclox (30µg), cefuroxime (30µg), ceftriazone (25µg), chloramphenicol (30µg). The zones of inhibition were measured and results recorded as sensitive (S) or resistance (R) based on NCCLs guidelines.

Plasmid Analysis

Plasmid DNA was isolated from cultured cells using alkaline lysis method as described by Birnboim and Doly (1979).

Plasmids were separated by electrophoresis in a 0.8% agarose gel.

Results

The prevalence of *Pseudomonas aeruginosa* and *Staphylococcus aureus* from burn wounds patients are as presented in Table 1. Of the 50 patients studied only 18 were infected by *Pseudomonas aeruginosa* and *Staphylococcus aureus*, with 16 (88.8%) being infected by *Pseudomonas aeruginosa* and 2 (11.1%) by *Staphylococcus aureus*. About 32 (64%) of hospitalized burn wound patients were not infected by *Staphylococcus aureus* or *Pseudomonas aeruginosa*.

Table 1: Prevalence of *Pseudomonas aeruginosa* and *Staphylococcus aureus* from burn wounds

Strain	Number
<i>Pseudomonas aeruginosa</i>	16 (88.8%)
<i>Staphylococcus aureus</i>	2 (11.11%)

Table 2: Antimicrobial susceptibility pattern

Isolates (n)	S	SXT	PEF	CN	AM	CPX	OFX	CH	SP	AU	E	APX	Z	R
<i>P. aeruginosa</i> (16)	16 (100)	16 (100)	11 (68.8)	16 (100)	16 (100)	8 (50)	13 (81.3)	16 (100)	13 (81.3)	16 (100)	-	-	-	-
<i>S. aureus</i> (2)	0 (00)	2 (100)	2 (100)	2 (100)	2 (100)	-	-	-	-	2 (100)	2 (100)	2 (100)	2 (100)	2 (100)

Key: S: Streptomycin, SXT: Spectrin, PEF: Pefloxacin, CN: Gentamycin
 Am: Amoxacillin, CPX: Ciprofloxacin, OFX: Ofloxacin,
 CH: Chloramphenicol, SP: Sparfloxacin, Au: Augumentin
 E; Erythromycin, APX: Ampiclox, Z: Cefuroxime, R: Ceftriazone
 - : Not tested

Table 3: Antibiotic resistance biogram of isolate

Isolate	Code No.	Resistance biogram	No. of resistance markers
<i>P. aeruginosa</i>	P ₁	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₂	Am, Au, CN, PEF, S, SXT, CH	7
"	P ₃	Am, Au, CN, PEF, OFX, S, SXT, CH, SP	9
"	P ₅	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₉	Am, Au, CN, PEF, OFX, S, SXT, CH, SP	9
"	P ₁₀	Am, Au, CN, S, SXT, CH, SP	7
"	P ₁₁	Am, Au, CN, S, SXT, CH	6
"	P ₁₂	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₁₃	Am, Au, CN, S, SXT, CH	6
"	P ₁₄	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₁₉	Am, Au, CN, OFX, S, SXT, CH, SP	8
"	P ₂₀	Am, Au, CN, OFX, S, SXT, CH, SP	8
"	P ₂₁	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₂₃	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₂₆	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
<i>S. aureus</i>	P ₂₈	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	10
"	P ₂₉	Am, Au, CN, S, SXT, CH, SP, CPX	9
"	P ₃₂	CPX, SXT, E, PEF, CN, APX, Z, Am, R	9

Table 4: Plasmid profiles of MDR *Pseudomonas* and *Staphylococcus* harbouring plasmids.

Isolates	Code No.	Resistance biogram	Plasmid size (bp)
<i>P. aeruginosa</i>	P ₁	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	≥ 1200
"	P ₂	Am, Au, CN, PEF, S, SXT, CH	-
"	P ₃	Am, Au, CN, PEF, OFX, S, SXT, CH, SP	-
"	P ₅	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	≥ 1200
"	P ₉	Am, Au, CN, PEF, OFX, S, SXT, CH, SP	"
"	P ₁₀	Am, Au, CN, S, SXT, CH, SP	"
"	P ₁₁	Am, Au, CN, S, SXT, CH	"
"	P ₁₂	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	"
"	P ₁₃	Am, Au, CN, S, SXT, CH	-
"	P ₁₄	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	≥ 1200
"	P ₁₉	Am, Au, CN, OFX, S, SXT, CH, SP	"
"	P ₂₀	Am, Au, CN, OFX, S, SXT, CH, SP	-
"	P ₂₁	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	≥ 1200
"	P ₂₃	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	-
"	P ₂₆	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	≥ 1200
"	P ₂₈	Am, Au, CN, PEF, OFX, S, SXT, CH, SP, CPX	"
<i>S. aureus</i>	S ₂₉	Am, Au, CN, S, SXT, CH, SP, CPX	-
"	S ₃₂	CPX, SXT, E, PEF, CN, APX, Z, Am, R	≥ 1200

Discussion

The rapid emerging of antibiotic resistance transfer gene among bacterial population are becoming important cause of clinical infection. These bacterial resistant genes are more vulnerable in debilitating patients especially those with immunosuppressed complication such as burns (Frier *et al.*, 1999; Prescott *et al.*, 2001).

The results indicates that *Pseudomonas aeruginosa* 16(88.8%) and *Staphylococcus aureus* 2(11.1%) could be implicated in burn wound. Bassak *et al.*, 1997 and Steer *et al.*, 1996, have also shown the association of *Pseudomonas* and *Staphylococcus aureus* in burn wound. The high prevalence of

Pseudomonas aeruginosa observed in hospitalized patients in this study is in accordance with those of Naghesha *et al.*, 1996.

An alarming increase in resistance of *Pseudomonas aeruginosa* and *Staphylococcus aureus* to various antimicrobial agents tested was observed in this study. *Pseudomonas aeruginosa* were all resistant to Streptomycin, Septrin, Gentamycin, Chloramphenicol and Augmentin. All *Staphylococcus aureus* were resistant to all antibiotic tested except Streptomycin (Table 2). Most reporters have also shown that *Pseudomonas aeruginosa* and *Staphylococcus aureus* are often difficult to treat due to high level of antibiotic resistance (Emori and Gyanes, 1993; Paul *et al.*, 1992). The study also revealed that all *Pseudomonas aeruginosa* and *Staphylococcus aureus* isolates from hospitalized burn wound patients in Central hospital, Warri harboured multiple drug resistance plasmids. Also remarkable in this study is the presence of similar plasmid size of $\geq 1.2\text{kpb}$, which suggests epidemiological relatedness. Sixty-one percent (61.1%) MDR isolates carried their resistance on plasmid. The plasmids molecular weight obtained in this study were quite small as compared to plasmids isolated by Aluyi and Arkortha, 2000, and Enabulele *et al.*, 1993.

MDR *Pseudomonas aeruginosa* and *Staphylococcus aureus* harbouring similar plasmid sizes were observed. These suggest that a single strain may be responsible for the transfer of MDR plasmids among these hospitalized burn wound patients. Therefore stringent measures directed against MDR should be enforced in hospitals.

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SCREENING OF THEILERIA PARVA APICOMPLEXAN ANTIGEN HOMOLOGS FOR INDUCTION OF MHC CD4+ AND CD8+ T-CELL RESPONSES

Nyerhovwo J. Tonukari^{1*} and Richard T. Kangethe²

¹Department of Biochemistry, Delta State University, PMB 1, Abraka, Nigeria.

²Department of Biochemistry, University of Nairobi, P.O. Box 30197, 00100, Nairobi, Kenya.

*Corresponding author E-mail: tonukari@gmail.com.

Abstract

Theileria parva, an intracellular apicomplexan pathogen transmitted by *Rhipicephalus appendiculatus* ticks, infects and transforms lymphocytes of cattle and African buffalo causing the disease called East Coast fever (ECF). The genome of *T. parva* was sequenced in order to facilitate research on parasite biology, assist the identification of schizont antigens for vaccine development and extend comparative apicomplexan genomics. In the present study, eight putative apicomplexan antigens were identified from literature and their homologs purified from a *T. parva* Muguga schizont cDNA library. These were immunoscreened for MHC CD4⁺ and CD8⁺ CTL response. The results showed that of the expressed proteins, only the *T. parva* T-complex 1 protein zeta subunit ortholog was found to elicit CD4⁺ response; none elicited CD8⁺ response. The elicitation of CD4⁺ T cell response by the *T. parva* T Complex Protein-1 zeta subunit homolog indicates that it is a candidate T-helper cell target antigen.

INTRODUCTION

Theileria parva, an apicomplexan pathogen causing economic losses to smallholder farmers in Africa, infects and transforms lymphocytes of cattle and African buffalo causing the disease called East Coast fever (ECF). Transmitted by *Rhipicephalus appendiculatus* ticks, the parasite causes a severe lymphoproliferative disease of cattle in eastern, central, and southern Africa (Katzer *et al*, 2006). It is an intracellular parasite that infects and transforms bovine lymphocytes. This disease, which kills over 1 million cattle each year in sub-Saharan Africa, results in economic losses exceeding \$200 million annually (Norval *et al*, 1992).

The genome of *T. parva* was sequenced in order to facilitate research on parasite biology, assist the identification of schizont antigens for vaccine development (Graham *et al*, 2006), and extend comparative apicomplexan genomics, in particular with *Plasmodium falciparum*, which causes malaria. The haploid *T. parva* nuclear genome is 8.3 x 10⁶ base pairs (Mbp) in length and consists of four chromosomes. Gardner *et al* (2005) also sequenced the parasite apicoplast and mitochondrial genomes (Kairo *et al*, 1994). The *T. parva* chromosomes contain one extremely A+T-rich region (>97%) about 3kbp in length that may be the centromere. The telomeric repeats are short. The *T. parva* nuclear genome contains about 4035 protein-encoding genes, 20% fewer than *P. falciparum*, but exhibits higher gene density, a greater proportion of genes with introns, and shorter intergenic regions.

Mining of sequence data has proved useful in the search for candidate vaccine antigens (Graham *et al*, 2006). Two approaches have been previously adopted for antigen identification in *T. parva* (Graham *et al*, 2006). Both techniques depend on screening of transiently transfected antigen-presenting cells with fully characterized cytotoxic T lymphocyte (CTL) (Taracha *et al*, 1995; Goddeeris and Morrison, 1988) from live vaccine-immunized cattle of diverse bovine leukocyte antigen (BoLA) major histocompatibility complex (MHC) class I genotypes. First, in a targeted gene approach, genes that were predicted by using preliminary sequence data from one of the four *T. parva* chromosomes (Gardner *et al*, 2005) to contain a secretion signal were immunoscreened. The approach was based on the observation that the schizont lies free in the host cell cytoplasm (Shaw, 2003) whereby signal peptide-containing parasite proteins would directly access the host cell MHC class I antigen processing and presentation pathway. In the second approach, a random immunoscreen of schizont cDNA clones was conducted because secretion of proteins that do not contain signal sequences has been reported (Nacer *et al*, 2001).

However, gene homologs encoding antigens from other apicomplexan parasites constitute a source of possible vaccine candidate antigens. This complimentary homolog screen approach to antigen identification would be to screen proteins with related amino acid sequence from related apicomplexan parasites that are known to be antigenic (Tonukari and Kangethe, 2009a,b). Here, eight putative apicomplexan antigens were identified from literature and their homologs purified from a *T. parva* Muguga schizont cDNA library (Graham *et al*, 2006). These were immunoscreened for MHC CD4⁺ and CD8⁺ CTL response.

MATERIALS AND METHODS

Cloning of targeted genes

The homologs of apicomplexan antigens described as antigenic in literature were selected for analysis (Tonukari and Kangethe, 2009b). The selected gene sequences were used to perform a BLAST search (Altschul *et al*, 1990; Altschul *et al*, 1997) of the *T. parva* genome database to identify homologous genes. Those identified in the *T. parva* database were translated to their amino acid sequences and analyzed using SignalP-2.0 and TMHMM software for the presence of signal peptides, glycosyl-phosphatidylinositol (GPI) anchor (which can be equated as a signal for their expression and possible antigenicity) and transmembrane domains. This would identify secreted proteins or proteins located on the surface of the parasite. Eight putative antigen identified (Table 1) were amplified from a purified *T. parva* Muguga schizont cDNA library (Graham *et al*, 2006) and cloned as previously described (Tonukari and Kangethe, 2009a,b).

PCR and cloning

Polymerase chain reaction (PCR) was performed with a thermocycler (MJ Research, Watertown, MA) using *Taq* DNA polymerase (Promega) and two primers based on the sequences identified in *T. parva* using cDNA library as template (Graham *et al*, 2006). The PCR product generated above was cloned into pGEM T-easy vector (Promega, Madison, WI). Vector specific primers, both forward and reverse were synthesized (Table 1). These primers were then used to amplify the ortholog genes from cDNA. All the PCRs were performed as described using the following conditions: initial denaturation at 94°C for 3 minutes, 35 cycles of denaturation, 94°C for 1 minute; annealing at 55°C for 1 minute and polymerization at 72°C for 2 minutes. A final round of polymerization at 72°C for 10 minutes was performed at the end of the 35 cycles. Aliquots (10µl) of PCR products with 5µl loading buffer were loaded onto a 0.8% TAE agarose gel, stained with ethidium bromide and visualized on a UV transilluminator.

PCR products were extracted and purified using QIA quick Gel DNA Extraction Kit protocol (QIAGEN Co.) and the purified PCR products ligated into pGEM-T Vector (Promega Co., USA) according to the manufacturer's instructions. 1µl of ligation reaction was transformed into *Escherichia coli* strain DH5-α competent cells. DNA nucleotide sequences were determined by gel based sequencing at the International Livestock Research Institute sequencing unit in Nairobi, Kenya. Sequences were analyzed using various basic alignment search tools (BLAST) served at the National Center for Biotechnology Information (NCBI) website (<http://www.ncbi.nlm.nih.gov/BLAST/>).

Expression and purification of *T. parva* proteins

After sequence verification, the cloned fragment or part of it was excised from pGEM-T by digestion with Hind III and XhoI and cloned into pET 28b, a bacterial based expression vector. The cloning sites used were generated by digesting with Hind III and XhoI. The constructs were transformed into *E. coli* BL21 (DE3). For expression using *E. coli* cells harbouring pET 28b with the cloned fragment, 500ml of 2xYT bacterial broth was inoculated with 50ml of overnight cultures of the respective *E. coli* transformed with homolog clones and grown in a 3 litre conical flask, in a 37°C rotatory incubator at 255rpm (Becton Dickinson, Franklin Lakes, NJ, USA) to an optical density of 0.6 at 600nm. Protein expression was induced by addition of isopropyl-β-D-thiogalactopyranoside (IPTG) to 2mM and incubation for 24 hr with time-lapse samples picked after 0, 3, 5 and 24 hrs. Bacterial cells were harvested, pelleted and weighed. Each batch of cells was then lysed with 5ml of 8M Urea, 0.1M phosphate buffer at pH 8.0 per 1g of pellet, by gently stirring overnight at 4°C. This was then spun down at 10,000g in a J100 Sorval

rotor to get rid of cell debris. The supernatant was then mixed with 2ml of 50% Ni-NTA agarose affinity matrix for 1 hr, and the resin washed three times with 8M urea-100mM Na₂HPO₄-50mM NaCl at pH 8.0 and once with the same solution at pH 7.5. Elution of bound recombinant proteins was performed by addition of 50mM ethylenediaminetetraacetic acid (EDTA). Eluted proteins were analyzed for purity and molecular mass on a 12.5% sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) gel, followed by Western blot analysis. The samples were then dialyzed against phosphate-buffered saline (PBS), passed through a 0.2µm filter, and protein concentrations determined by a bicinchoninic acid protein assay (Pierce).

Western blot analysis

Expressed and purified proteins were confirmed using western blotting. 5µg of each sample was run on a 12.5% SDS-PAGE gel at 35mA in a 1 X running buffer (25mM Tris, 250mM glycine (electrophoresis grade) pH 8.3, 0.1% SDS prepared from a 10x running buffer stock solution). The transfer of proteins onto nitrocellulose sheets was performed as described by Towbin *et al.* (1979). After electrophoresis, the gel was equilibrated in transfer buffer (10% methanol, 24mM Tris, 194mM glycine) for 30 minutes to avoid any change in its size during transfer. The samples were electro-transferred onto nitrocellulose paper (0.45µm protan nitrocellulose, Schelcher and Schuell, Dassel, Germany) at 70V for 1 hr at 4°C, or 15V overnight, in transfer buffer. The blot was then stained with ponceau S (Sigma Aldrich, St. Louis MO, USA) to monitor the transfer, and then destained in transfer buffer and several rinses with water. The nitrocellulose filter was then blocked with blocking solution (TBS-Tween: 20mM Tris-HCl, 200mM NaCl, pH 7.4 containing 5% non-fat dry milk) for 1 hr. The primary antibody, anti-His-tag protein (His-tag monoclonal antibody, Novagen, EMB Sciences, SanDiego CA, USA), was added to the blocking solution at a dilution of 1:2000 with 0.02% NaN₃ and incubated overnight. The next morning the filter was washed 4 times with PBS/0.1% Tween 20, 10 minutes each. The second antibody, rabbit anti-mouse IgG HRP conjugate (Amersham International PLC, Aylesbury Buckinghamshire, England), was added at a dilution of 1:1500 in blocking solution and incubated for 3 hr. The filter was then washed two times with PBS/0.1% Tween 20, then twice with PBS. The blot was developed by addition of the chromogenic substrate 3, 3-diaminobenzidine (DAB; Sigma) in PBS at a concentration of 1mg/ml in the presence of 0.1% (v/v) hydrogen peroxide. The filter was rinsed rapidly with water and dried sandwiched between two Whatmann 3mm filters.

SDS-PAGE

Total proteins as well as the purified *T. parva* proteins from the *E. coli* cells harbouring pET 28b with the cloned fragment were analysed by SDS-PAGE according to Laemmli (1970) using 12% polyacrylamide gels followed by staining with coomassie brilliant blue. Protein quantitative analysis was determined by the Bradford method (Bradford, 1976) with bovine serum albumin as a standard.

Transfection of iSF and COS-7 Cells

iSF were immortalized by using standard procedures (Jha *et al.*, 1998) with modifications (unpublished observations). iSF were transfected in 96-well plates with clones or pools of schizont cDNA (100ng per well) by using FuGENE 6 (Roche Diagnostics, Mannheim, Germany) and cultured for 24 hr. COS-7 cells were cotransfected with 100ng of each clone per well or pooled together with 50ng of pcDNA3 BoLA-N*00101 (Bensaid *et al.*, 1991) or BoLA-N*01301 cDNA per well (Graham *et al.*, 2006).

IFN-γ ELISpot

Transfectants were cocultured with schizont-specific CTL, and recognition was assessed by using an IFN-γ ELISpot assay (Taracha *et al.*, 2003; Graham *et al.*, 2006).

RESULTS AND DISCUSSION

Eight *T. parva* homologs of apicomplexan antigens were isolated and sub-cloned. Recombinant proteins were generated using *E. coli* expression vector system. The expressed proteins were tested by ELIspot. Only the *T. parva* T-complex 1 protein zeta subunit ortholog was found to elicit CD4⁺ response; none elicited CD8⁺ response (Table 2; Figure 1).

The *T. parva* ortholog of the zeta subunit of T-complex protein 1 (TCP-1) which plays a role in protein folding, assembly and transport has been previously reported (Tonukari and Kangethe, 2009a). The deduced amino acid sequence of the *T. parva* ortholog has a 55% identity and 77% similarity to the same protein in a number of eukaryotes such as *Danio rerio* (zebra fish) and man. T-complex protein I is a chaperonin-containing protein (Kubota *et al*, 1995) and is abundant in the eukaryotic cytosol. It is involved in the folding of actin and tubulin concomitant with ATP hydrolysis *in vitro*. One of the characteristics that distinguishes TCP-1 from other chaperonins is its hetero-oligomeric nature, which in general comprises of eight different polypeptide species (Kubota *et al*, 1994).

The major mechanism responsible for the elimination of cells infected with the parasite is MHC-class 1-restricted (parasite specific) CD8⁺ CTL activity. However, CTL activity requires the input of specific CD4⁺ T-cell help for induction. Katzer *et al*. (2006) observed that protective immunity against *T. parva* has considerable impact on the emergence of targeted parasites following challenge but fails to prevent their differentiation into transmissible forms. Furthermore, the genotypic compositions of the transmitted parasites arising from a given challenge vary between immune individuals with distinct MHC phenotypes.

Five candidate vaccine antigens that are the targets of MHC class I-restricted CD8⁺ CTL from immune cattle have been previously identified (Graham *et al*, 2006). Schizont-infected cell-directed CD8⁺ cytotoxic T lymphocytes (CTL) constitute the dominant protective bovine immune response after a single exposure to infection. CD8⁺ T cell responses to these antigens were boosted in *T. parva*-immune cattle resolving a challenge infection and, when used to immunize naïve cattle, induced CTL responses that significantly correlated with survival from a lethal parasite challenge. These data provide a basis for developing a CTL-targeted anti-East Coast fever subunit vaccine (Graham *et al*, 2006).

According to Graham *et al*, (2006), the ultimate vaccine against *T. parva* will almost certainly need to incorporate multiple antigens and epitopes in order to confer protection in the genetically diverse outbred cattle population exposed to challenge by antigenetically diverse parasite populations in the field. Furthermore, the major challenge in the development of a vaccine against *T. parva* based on the induction of T-cell responses will be to generate responses that are effective against all parasite strains. The elicitation of CD4⁺ T cell response by the *T. parva* T Complex Protein-1 zeta subunit homolog indicates that it is a candidate T-helper cell target antigen. Incorporation and further evaluation of this antigen for vaccine design in East Coast fever disease should be of interest.

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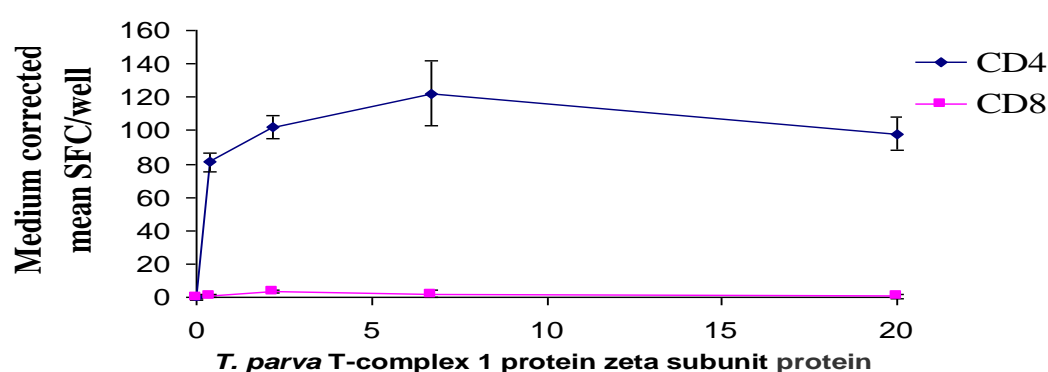


Figure 1 (colour online): Response of *T. parva* specific CD4⁺ and CD8⁺ polyclonal T cells to recombinant *T. parva* T-complex 1 protein zeta subunit protein. *T. parva* specific CD4⁺ and CD8⁺ polyclonal T cell lines isolated from ITM immunised cattle BW014 were co-cultured with autologous monocytes and stimulated with a titration of truncated recombinant *T. parva* T-complex 1 protein zeta subunit protein. Responses are presented as mean numbers of spot-forming cells (SFC)/well.

Table 1. Primers for amplifying *T. parva* apicomplexan homologs.

Putative antigen	Apicomplexan parasite	Primer name	Primer sequence
12D3 antigen	<i>Babesia</i> spp.	Forward	GCCGCCACCATGGGCTACTACCCACTTCCTTCAAC
		Reverse	TTAAAGTTCATTACACGTTTGAACACTT
110kDa antigen	<i>Plasmodium knowlensi</i>	Forward	GCCGCCACCATGAAAGATGAAGAGTTGCATCATGC
		Reverse	TTAACAAGTTACTTCCGTTCCGCTCTGA
T-complex protein1	<i>Plasmodium falciparum</i>	Forward	GCCGCCACCATGTCTGAAGAGTATGCCATCTATTGA
		Reverse	TTACTCTGTACCAATTAGCGTAACTTGA
Apical membrane antigen (AMA1)	<i>Plasmodium chabadi</i>	Forward	GCCGCCACCATGAGTTTTAGCCCTAACACTGCTGA
		Reverse	TTATATGAATGGTCCTGAAGAAACGGGT
22.4.1 protein	<i>Cryptosporidium parvum</i>	Forward	GCCGCCACCATGAGTTCAAGGTATAAAAGAAATTA
		Reverse	TTAGGAGGGTGTGTATTCAGGGCAGTTG
T-complex protein zeta subunit	<i>Plasmodium falciparum</i>	Forward	GCCGCCACCATGGCAGTCAATATCTTAAATAGCAG
		Reverse	TTACGAAGGAGCGTTATGCATAGACCTT
T-complex protein 1, delta subunit	<i>Babesia microti</i>	Forward	GCCGCCACCATGCCACCGCCTTCTAATAATTCTGT
		Reverse	TTACTCGAACATGTCAGAGATGCTATTC
Ring-infected erythrocyte surface antigen	<i>Plasmodium falciparum</i>	Forward	GCCGCCACCATGTCCGAATGTGATACCATGGAGAT
		Reverse	TTAGTAAAAGGGCTCGTTTGAGTAGTGT
HSP-70	<i>Plasmodium berghei</i>	Forward	GCCGCCACCATGAGTTGCATTTTAAAGTGTAATGA
		Reverse	TTAATTTTGATCATTATTATTATCAATG

Table 2. Apicomplexan antigen homologs identified in *Theileria parva*.

Putative antigen	Apicomplexan parasite	<i>T. parva</i> homolog ORF (bp)	<i>T. parva</i> homolog CD4 ⁺ response	<i>T. parva</i> homolog CD8 ⁺ response
12D3 antigen	<i>Babesia</i> spp.	1218	—	—
110kDa antigen	<i>Plasmodium knowlensi</i>	936	—	—
T-complex protein1	<i>Plasmodium falciparum</i>	489	—	—
Apical membrane antigen (AMA1)	<i>Plasmodium chabadi</i>	1425	—	—
22.4.1 protein	<i>Cryptosporidium parvum</i>	576	—	—
T-complex protein zeta subunit	<i>Plasmodium falciparum</i>	1647	+	—
T-complex protein 1, delta subunit	<i>Babesia microti</i>	1479	—	—
Ring-infected erythrocyte surface antigen	<i>Plasmodium falciparum</i>	1710	—	—
HSP-70	<i>Plasmodium berghei</i>	792	—	—

EFFECT OF BARBADENSIS MILLER SPP PLANT EXTRACT ON RATS INDUCED WITH HEPA TIC INJURY

Kadiri E. Helen

DEPARTMENT. OF BIOCHEMISTRY, DELTA STATE UNIVERSITY, ABRAKA.

E-mail: hekad@yahoo.com

Abstract

Forty male albino rats (Wistar Strain) weighing between 120 and 170g were used to study the antioxidant property of *Barbadensis miller* (Aloe Vera extract). The rats were divided into 4 groups each group consisting of 10 animals. The antioxidant activity of the extracts were evaluated using ccl_4 induced lipid peroxidation model. Group one was kept on normal diet and served as control, the second group received the extract alone three times daily for 10 days by oral route, the third received only ccl_4 in olive oil by subcutaneous injection, while the fourth group received the extract at the same dose and duration as group two, before exposure to ccl_4 . Eighteen hours after ccl_4 administration, the animals were sacrificed, blood was collected and serum separated for analysis. Biochemical analysis of serum indicate increased activities of alkaline phosphatase (ALP), serum glutamic oxaloacetate transaminase (SGOT) and Glutamic pyruvic transaminase (SGPT) and serum concentration of bilirubin in ccl_4 administered rats which is indication of liver damage occasioned by lipid peroxidation. Prior treatment of ccl_4 exposed rats with the plant extract lowered the serum activation of these enzymes to levels that were comparable to control. The study indicates that aqueous extract of *Barbadensis miller* spp shows antioxidant property since it improves recovery or reduces the toxic effects of ccl_4 in liver cells of male rat.

Keywords: *albino rats, Barbadensis miller spp., ALP, SGOT and SGPT.*

INTRODUCTION

The liver is vulnerable to a wide range of metabolic, toxic, microbial, circulatory, and neoplastic insults. The level and the state of injury to the hepatocyte can be ascertained by various markers. The most commonly used markers of hepatocyte injury L-aspartate aminotransferase and L-alanine aminotransferase. Although, if hepatic disease is primary of an obstructive nature (cholestatic), alkaline phosphatase will be significant enzyme marker. It has been shown that hepatic injury or disease results in the leakage of these enzymes into circulation thereby increasing their concentration (Uliena *et al.* 2003). However the pathogenesis of induced hepatic injury is not very clear, however there is no doubt that Reactive Oxygen Species (ROS) and free radicals play an important role in biochemical changes taking place in liver. Biochemical membranes are particularly prone to the effect of ROS. The peroxidation of unsaturated fatty acids in biological membranes lead to decrease in membrane fluidity and a disruption in membrane integrity and function which may cause serious biochemical changes.

Carbon tetrachloride (CCl_4) is a hepatotoxin. Trichloromethyl radicals are generated from it. The radicals stimulate a sequence of reactions that culminate in the initiation of the peroxidation of membrane lipids (Reinke *et al.*, 1988.) and hence liver damage. Trichloromethyl radical is believed to be the immediate product of the reductive dechlorination of CCl_4 catalysed by certain cytochrome P₄₅₀ isoenzymes (Sipes *et al.*, 1977) several endogenous protective mechanisms have been evolved to limit ROS and the damage caused by them. (Uliena *et al.*, 2003), but this may not provide complete protection from oxidative stress. Hence, there is increased need of natural and artificial agents possessing artificial properties.

Aloe vera is a medicinal plant that belongs to the family Liliaceae. It is indigenous in dry regions of Africa, Asia, Europe and America. The *Bardadensis* Miller species of Aloe vera contains minerals essential to human and animals. Vitamins A, B, B₂, B₆, B₁₂, C and E as well as amino acids. Aloe vera has a wide variety of properties and activities. In relatively small concentrations together with the gel fraction, they may provide acclaimed analgesic, antibacterial, antifungal and antiviral activity. It is also used as a laxative due to the anthraquinones in it. The antioxidant activity of vitamin A, C and E in Aloe vera makes it a good hepatoprotector in CCl₄ – induced liver injury. The vitamins neutralizes free radicals thereby inhibiting peroxidation of unsaturated fatty acids in biological membranes.

MATERIALS AND METHODS

Materials

Forty male albino rats, Wistar strain (150 – 170g) bred in the animal house of the college of medicine, University of Lagos, Nigeria were used for the study after permission from appropriate authorities. They were divided into four experimental groups with ten animals per group. The animals were left to acclimatize to laboratory conditions for two weeks, before the commencement of the study.

L-alanine/L-aspartate amino transferase kits were products of Quimica clinica Applicada (QCA), Spain. Para-nitrophenol phosphate and para-nitrophenol were purchased from BDH chemicals (Poole, England). Tartaric and citric acids were obtained from Merck chemicals (Darmstadt). Grower's mash was obtained from Livestock Feeds Plc., Nigeria. The fresh plant of Aloe vera were obtained from a farm in Abraka, Nigeria and authenticated by the department of Botany, Delta State University, Abraka, Nigeria.

Preparation of Aqueous Extract of Aloe vera

The aqueous extract was prepared by boiling 100g of the plant in 1000 mL of water for 15min with subsequent standing to allow for cooling down to room temperature. After separation of undissolved residue, the solution was used for the study.

Treatment of Animals

Rats in group 2 and 4 were given 5mL of extract kg⁻¹ body weight orally by incubations. At the same time animals in group 1 and 3 received an equal volume of de-ionized water kg⁻¹ body weight by the same route. This treatment was carried out three times daily for ten days during which the rats were allowed free access to food and water. Following the last treatment, rats in group 3 and 4 received a mixture of CCl₄ in olive oils (1:1) subcutaneously at a dose of 6 mL kg⁻¹. The CCl₄- free control rats (group 1 and 2) were given 6 mL olive oil kg⁻¹ body weight subcutaneously. Treatment of the animals was in accordance with the principle of laboratory animal care (NIH, 1985).

Preparation of Serum

Eighteen hours after CCl₄ treatment, each rat was anaesthetized in a chloroform saturated chamber. The thoracic and abdominal regions were opened to expose the heart. Blood was obtained through heart puncture by means of a 5 mL hypodermic syringe and needle and placed in ice-cold 10 mL centrifuge tube. It was allowed to clot and then centrifuged at 3000 g for 5 min. The serum sample was collected and left standing on ice until required.

Biochemical Assays

Serum aspartate and alanine aminotransferases activities were assessed using Quimica Applicada kits based on the methods of Reitman and Frankel (1957). The activities of L-ALT and L-AST are expressed as units mL⁻¹. Alkaline phosphatase activity was determined, by the method of Annino and Giese (1976). The enzyme activities expressed in units L⁻¹ in which one unit represent one micromole p-nitrophenol produced per minute.

Statistics

The results are expressed as mean±SEM and the mean values of the groups were compared using ANOVA and least-square difference. The significance level was set at p<0.05.

RESULTS

It was observed that the size of the liver was increased in CCl₄-intoxicated rats, but it was not significantly different from the control in rats both treated with extract alone and those pretreated with extract before CCl₄ administration. The body weight gain of rats did not differ significantly between the groups. Thus the study indicates that aqueous extracts of Aloe vera restored to normal the CCl₄ induced increase in liver/body weight ratio of rats (Table i)

Table ii presents the effect of Aloe vera extract on liver function indices of CCl₄ treated rats. The activities of serum aminotransferases (L-ALT and L-AST) of rats administered CCl₄ (group 3) and extract alone (group 2) were significantly increased relative to control. Prior administration of the extract to CCl₄ treated rats (group 4) decreased the activities of the serum aminotransferases to levels that were similar to the control. Similarly the serum alkaline phosphatase activity was significantly increased in CCl₄ treated extract-free rats as compared to control and extract treated rats.

Table i. Effect of Aloe vera extract on liver/body weight gain of CCl₄ treated rats

	Group 1	Group 2	Group 3	Group 4
Parameters	-CCl₄ – ext	-CCl₄ + ext	+CCl₄ -ext	+CCl₄ + ext
Liver/body weight ratio(10⁻²)	2.50 ± 0.38 ^a	2.40 ± 0.20 ^a	5.40 ± 0.30 ^b	2.58 ± 0.40 ^a
Body weight gain (g/rat⁻¹day⁻¹)	3.70 ± 0.20 ^a	3.30 ± 0.20 ^a	3.20 ± 0.59 ^a	3.60 ± 0.50 ^a

Values are mean ± SEM, values on the same row with different superscript differ significantly (P<0.05) from each other

Table ii.

Effect of Aloe vera extract on serum L-aspartate aminotransferase (L-AST), L-alanine aminotransferase (L-ALT) and alkaline Phosphatase (ALP) activities of CCl₄ Treated rats

	Group 1	Group 2	Group 3	Group 4
Parameters	-CCl ₄ - ext	-CCl ₄ + ext	+CCl ₄ -ext	+CCl ₄ + ext
AST (units mL ⁻¹)	162 ± 2.50	163.20 ± 2.0	180.10 ± 1.50	165.15 ± 2.00
ALT (units mL ⁻¹)	142 ± 1.50	139.35 ± 1.20	166.30 ± 3.80	145.18 ± 0.85
ALP (units L ⁻¹)	50.70±2.46	48.15 ± 3.80	72.25 ± 2.65	53.21 ± 0.125

However pre treatment of CCl₄, administered rats with the extract significantly decreased the activity of alkaline phosphatase relative to CCl₄ treated extract-free rats. Therefore the study indicates that the extract protects rats from CCl₄, - induced liver damage.

Direct change in organ weight or organ /body weight ratio has been used as an index of CCl₄, toxicity (Uemitsu *et al.*, 1986; Uemitsu and Nakayoshi, 1984). The latter method was used in this study since it has been shown to be a more sensitive indicator of CCl₄, toxicity than absolute liver weight (Uemitsu *et al.*, 1986). Therefore the observed increase in liver/body weight ratio of rats administered CCl₄, (Table i) is consistent with these reports and that of others (Vilstrup, 1983; Okazaki *et al.*, 1985). Increase in organ weight after exposure to a toxicant may be due to a tumor, fluid or triglyceride accumulation. Triglyceride accumulation in liver (fatty liver) is a common response to CCl₄ toxicity (Timbrell, 1991; Junnila *et al.*, 2000) and this may account for the observed increase in the liver/body weight ratio of the CCl₄, -treated rats. In this study, the hepatoprotective activity of Aloe vera extract in CCl₄, -induced toxicity was evaluated using changes in liver body weight ratio and body weight gain of rats since they are well known indices of CCl₄ toxicity. The reversal of the CCl₄, -induced increase in liver/body weight ratio of rats by the extract is an indication of its protective effect.

Serum aminotransferases (ALT and AST) and serum alkaline phosphatase activities were also used to measure both CCl₄, -induced hepatotoxicity and protection of Aloe vera extract against the same effect of CCl₄, in rats. In agreement with previous reports (Reinke *et al.*, 1988), our results show that CCl₄ caused an elevation in the serum levels of ALT, AST and ALP which is indicative of damage to the liver. Treatment of rats with aqueous extract of Aloe vera caused less hepatotoxicity than with CCl₄, alone (Table ii) as evidenced by the decreased serum content of the above enzymes relative to the CCl₄, -treated extract-free group. Various mechanisms have been proposed for CCl₄, -induced liver, damage (Brattin *et al.*, 1985). One view is that a trichloromethyl radical (CCl) is produced from CCl₄ by reductive dechlorination.. The trichloromethyl radical in turn abstracts a hydrogen atom from a fatty acid to form chloroform and a lipid radical. The lipid radical may then react with molecular oxygen to initiate lipid peroxidation which is thought to ultimately cause the cytotoxic response (Sipes *et al.*, 1997 Recknagel, 1983; Brattin *et al.*, 1985; Reinke *et al.*, 1988). In the present study, we did not attempt to address any mechanistic concept but merely adopted CCl₄, -induced liver injury as model for accessing Aloe vera for antioxidant activity. Since the mechanism of action of CCl₄ involves oxidation, we are of the view that since Aloe vera possesses antioxidant action it would prevent lipid peroxidation and therefore membrane damage. Present result clearly demonstrate that Aloe vera is a good preventive agent for CCl₄, -induced liver damage in rats.

Many natural and artificial agents possessing anti-oxidative properties have been proposed to prevent and treat hepatotoxicity induced by oxidative stress (Lieber, 1997.; Eervincova and Drahota, 1998) There is increasing evidence for the hepatoprotective role of hydroxyl- and polyhydroxyl organic compounds particularly from vegetables, fruits and some herbs (Bass, 1999). Although the biochemical mechanism of the antioxidant effect of Aloe vera extract against CCl₄, -induced hepatotoxicity was not examined in the present study, it is possible that the bioactive principles in the extract may have acted

directly or indirectly in protecting the liver against damage. Directly it may be breaking the sequence of events between the reductive dechlorination of CCl_4 and the subsequent abstraction of hydrogen from unsaturated fatty acids in the membrane and peroxide formation. Indirectly, it may inhibit the activities of cytochrome P_{450} isoenzymes (Sipes et al., 1977; Reinke et al., 1988) required for trichloromethyl radical formation or it may be an effective scavenger of the reactive metabolite, the trichloromethyl radical. Besides the increase in serum L-ALT and L-AST of rats exposed to the extract alone, (Table ii) may arise following increased synthesis of these enzymes in the liver. Activation of aminotransferases may promote regeneration of damaged hepatocytes and this could contribute to the restoration of CCl_4 -induced liver damage by prior treatment of rats with the extract.

CONCLUSION

In conclusion the present study indicates that aqueous extract of Aloe vera has anti-oxidant activity since it improves recovery or reduces the toxic effects of CCl_4 in liver cells of male rats.

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PHYTOCHEMICAL SCREENING OF TEN NIGERIAN LOCAL MEDICINAL PLANTS

Akpovwehwee A. Anigboro

Department of Biochemistry, Faculty of Science, Delta State University, Abraka, Nigeria .

Abstract

Phytochemical screening of ten selected Nigerian medicinal plants was investigated. The plants investigated are: *Achyranthes aspera*, *Acalypha godseffiana*, *Ocimum gratissimum*, *Capolobia lutea*, *Senna alata*, *Anthocheista djalensis*, *Blighia sapida*, *Enantia chlorantha*, *Chromolaena odorata*, and *Nauclea latifolia*. The phytochemicals that were determined qualitatively and quantitatively were: saponins, alkaloids, phenols, and flavonoids. The presence of saponins, alkaloids, phenol and flavonoids were observed in all the investigated plants. The percentage amount of saponins ranges from was highest in *E. chlorantha* (6.00 ± 0.12). The percentage amount of alkaloids was highest in *A. godsettian* (1.03 ± 0.60), phenols (1.83 ± 0.14) and flavonoids (1.30 ± 0.01) were highest in *C. odorata*. These plants are important in medicine, in pharmaceutical companies and in food industries.

Key words: Phytochemical screening, Saponin, Alkaloids, Phenols, Flavonoids and Medicinal Plants.

INTRODUCTION

Nature has been a source of medicinal agents since times immemorial. The importance of herbs in the management of human ailments cannot be over emphasized. It is clear that the plant kingdom harbors an inexhaustible source of active ingredients valuable in the management of many intractable diseases. Furthermore, the active components of herbal remedies have the advantage of being combined with many other substances that appear to be inactive. However, these complementary components give the plant as a whole a safety and efficiency much superior to that of its isolated and pure active components. (Shariff, 2001).

Largely as a consequence of their sessile life styles, plants accumulate hundreds of thousands of specialized compounds called phytochemicals primarily derived from secondary metabolism (Grotewold, 2001). Phytochemicals are non-nutritive plant chemicals that have protective or disease prevention properties. These phytochemicals are often synthesized in cellular compartments different from where they accumulate. Many phytochemicals are secreted, either in a constitutive fashion, or in response to specific biotic or abiotic conditions (Alker, 2003).

It is well known that plants produce these chemicals to protect themselves, but recent research demonstrates that they can protect humans against diseases. Some of the well known phytochemicals are cardiac glycosides, flavonoids, steroids, tannins, saponins and alkaloids. The screening of plant extracts and plant products has shown that higher plants represent a potential source of novel antibiotic prototypes (Afolayan, 2003). Recent work revealed the potential of several herbs as sources of drugs (Iwu, 2002). Antibiotic resistance has become a global concern (Westh et al., 2004). There has been an increasing incidence of multiple resistances in human pathogenic microorganisms in recent years, largely due to indiscriminate use of commercial antimicrobial drugs commonly employed in the treatment of infectious diseases. This has forced scientists to search for new antimicrobial substances from various sources like the medicinal plants. These anti-nutritional factors are also known as 'secondary metabolites' in plants and they have been shown to be highly biologically active (Zank, 1991). Although most of these secondary metabolites elicit very deleterious biological responses, some of them have found a wide application in

nutrition and as pharmacologically active agents (Oakenfull and Sidhu, 1989). Saponins and flavonoids, for example, have found wide applications in the fields of medicine, pharmacy and food industries as pharmacologically active principles (Schopke and Hiller, 1990); in food, drink and beverage industries as foaming agents (Fenwick et al., 1983; Oakenfull and Sidhu, 1989), as antioxidants, preservatives and flavouring agents (You et al., 1993; Fenwick et al., 1983) and in agriculture especially as allelochemicals (Oleszec et al., 1992; Waller et al., 1993). In the native African traditional medicine and folk medicine, concoctions prepared from plant materials using water or local alcohol may contain a complex mixture of hundreds of bioactive plant secondary metabolic constituents (Igile, 1995). The nutritional, biochemical and physiopathological effects of these anti-nutritional factors have been reviewed by Aletor (1993).

The aim of this research is to highlight the pharmacological and other applications of these secondary plant metabolites and to emphasize on the need for them to be phytochemically exploited as phytomedicines and functional foods.

This study investigates the fundamental scientific bases of the use of some medicinal plants by defining and quantifying the percentage of crude phytochemical constituents present in these plants. The ethnobotanical information of some of the Nigerian traditionally used plant species selected for the phytochemical screening are as shown in Figures 1-10.

MATERIALS AND METHODS

Identification of Plant Materials

All the ten plant samples were completely identified in the Department of Botany, Delta State University, Abraka, Nigeria.

Collection of Plant Material

The leaves of the plants screened in this investigation were collected from different localities including uncultivated farmlands located at Abraka, Delta state, Nigeria. The collection of these commonly occurring native plants was done during the flowering period in wet season.

Preparation of Plant Extract

After the collection and identification of plant samples, the plant samples were dried in room temperature and ground into uniform powder using a Blender Mill Grater. The aqueous extract of each sample was prepared by soaking 100 g of dried powdered samples in 200 ml of distilled water for 12 h. The extracts were filtered using Whatmann filter paper No 42 [125mm].

Phytochemical Screening

Chemical tests were carried out on the aqueous extract using standard procedures to identify the constituent as described by Sofowara (1993), Trease and Evans (1989), Harbone (1973) and Edeoga et al. (2005).

DISCUSSION

Ten Nigerian medicinal plants were analyzed qualitatively and quantitatively for the presence of saponins, alkaloids, phenols, and flavanoids as shown in tables 1 and 2, respectively. The presence of saponins, alkaloids, phenol and flavonoids was observed in all the investigated plants. The results of the qualitative analysis of these plants showed that the plants (10) contained all the parameters (saponins, alkaloids, phenol and flavonoids) analyzed in varying proportion according to the intensity of colour. The degree of the presence of these parameters are indicated by mild (+), moderate (++), intense (+++) and absence (-). Hence, the degree of colour intensity can be used to evaluate the quality of the parameters above. The results of the quantitative analysis revealed that saponins content was highest when compared to other parameters analyzed. The saponins content was highest in *E. chlorantha* 6.00 ± 0.12 and lowest in *O. gratissimui*. Although different amounts of flavonoids content were present in the different plants, flavonoids content was highest in *A. djalonsensis* (1.32 ± 0.01) and lowest in *A. aspera* (0.10 ± 0.01).

Percentage crude alkaloids content varied with the plants and *A. godsetijana* had the highest (1.03 ± 0.60). The percentage content of crude phenol also varied with the plants and was highest in *C. odorata* (1.83 ± 0.14).

The % crude yield of flavonoid content of the plants studied showed that the leaves had high levels of flavonoids. A similar result has been reported by Sofowara (1993) that most plants are known to show medicinal activity as well as exhibiting physiological activity. *A. djalensis* possessed very high levels of flavonoids and are employed in the treatment of Bronchial ailment, mental disorders, skin diseases, abdominal pain, cancer and other infectious diseases. This is in accordance with strong experimental evidence that flavonoids possess the inherent ability to modify the body's reaction to allergens, viruses and carcinogen. They show antimicrobial and anti-inflammatory activity (Lamb and Cushine, 2005). A similar result of the presence of flavonoids in *A. godsettiana* has also been reported (Egunjobi, 1969, Owolabi et.al., 2008, Edwaga and Gomena, 2002). The flavonoid content of these medicinal plants can be useful in industrial processes. This is evident in the finding of Zviak and Charles 1986, which indicated that certain flavonoids are known for their use in the preparation of cosmetics composition as agents for protecting the skin and its superficial growth from lighting. The flavonoids content of medicinal plants has various pharmaceutical properties, including antioxidant, bactericidal and antiviral (Sahelian, 2005).

In the chemical industry, flavonoids are used in the manufacture of insecticides using the isoflavonoid, rotenone (Harborne, 1967) and in the preparation of various cosmetic products, where they are used as natural stabilizers and preservatives and are synergistically used to enhance the antimicrobial activities of many skin lotions and their products. Flavonoids suppress the effects of active oxygen species (H_2O_2 and O_2) in many other vulnerable biological systems (Nakayama et al., 1993). Flavonoids are used as natural anti-oxidants in food, and non -nutritive plant materials due to their ability to inhibit and scavenge reactive oxygen species (Kim et al., 1990; Larson, 1988).

The medicinal importance of tannins, saponins, which are components of traditional herbal preparation used in managing various common ailments, has been reported by Addae – Mensah (1992).

The importance of saponin in various antibiotics used in treating common pathogens has been recently reported by Kubumarava (2007). The beneficial effects of saponins are largely due to their hypocholesterolaemic action, leading to the belief that they may prove useful in the control of human cardiovascular disease (Oakenfull and Sidhu, 1983). The hypocholesterolaemic activity of dietary saponins may be due to the formation of some complexes with dietary cholesterol or their bile salt precursors which can then be made unavailable for absorption.

Johnson et al. (1986) reported that besides lowering serum cholesterol, saponins also readily increase the permeability of the mucosal cells of the small intestine, thereby facilitating the uptake of materials to which the gastrointestinal tract would not normally be permeable. Certain plant extracts which contain saponins are used as flavourings in food (Merck, 1960) and as foam producing agents. Purified saponins or their concentrated extracts are used as food additives in the manufacture of food and drinks primarily as foaming agents or as emulsion stabilizers. Saponins are also used as an anti-oxidant for food use (Takashi et al., 1986). Saponins are used in the preparation of spray dried powders containing vitamin E for the enrichment of foods, drinks and animal feeds.

The results of the quantitative analysis showed that phenol was present in all the plants investigated. *C. odorata* (Shell leaf) contained highest mean values (1.831 ± 0.14) in percentage of crude extract. Hence their high medicinal value may probably account for their use in the treatment of wounds, fever, cough and diabetes (Gordian et al., 2007). *A. aspera* also contained high amounts of phenol; it has not been previously reported in literature for their phenolic composition and their medicinal potency.

The use of the following plant *Ocimum gratissimum*, in the management of fever and other ailments has been reported (Burkell, 1984; Gill, 1992; Agolia, 1981; Egunjobi, 1969; Holms et.al., 1997).

Similar result for the presence of alkaloids in *E. chlorantha* has been reported (Back house et.al, 1994).

Also, the result of the quantitative analysis presented in Table 2 showed that alkaloid was present in all plants investigated. In all the plants investigated *A. godsettiana* (1.03 ± 0.60), contained highest mean values in percentage of crude extract, hence their medicinal value may probably account for their use in the treatment of fever and other ailment (Burkell, 1984; Gill, 1992; Agolia, 1981; Egunjobi, 1969; Holms et.al., 1997).

Conclusion

This study showed that there are a lot of phytochemicals present in Nigerian medicinal plants which can be exploited for the treatment of certain ailments. They can also be employed in pharmaceutical companies as well as in food industries. My future research will be to investigate the effect of some of these plants on some blood and organ parameters in experimental animals. Their antimicrobial effects shall also be investigated.

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Table 1: The results of the qualitative analysis of flavonoids, saponins and Alkaloids, in 10 Nigeria local medicinal plants.

S/N	Plants (Botanical names)	Flavonoids	Saponins	Alkaloids
1	<i>Achyranthes aspera</i> **	+	+	+
2	<i>Ocimum gratissimum</i> **	++	+	++
3	<i>Acalypha godsettiana</i> **	+	+	+
4	<i>Capolobia lutea</i> **	+++	+	+
5	<i>Chromolaena odorata</i> **	++	+	+
6	<i>Senna alata</i> **	++	+	+
7	<i>Nauclea latifolia</i> **	++	+	+
8	<i>Anthocheista djalensis</i> **	++	+	+
9	<i>Blighia Sapida</i> **	++	+	+
10	<i>Enantia chlorantha</i> **	+++	+	+

Table 2. The results of the quantitative determination of the % crude phenol, flavonoids, saponins and alkaloids.

S/N	Plants (botanical names)	% Crude extract of phenol	% Crude extract of flavonoid	%Crude extract of saponins	%Crude extract of alkaloids
1	<i>A. aspera</i>	0.96±0.25	0.10±0.01	3.79±0.20	0.206 ± 0.27
2	<i>O. gratissimum</i>	0.33±0.10	0.49±0.01	0.00±0.00	0.717 ± 0.50
3	<i>A. godsettiana</i>	0.81±0.10	0.99±0.01	4.03±0.15	1.030 ± 0.60
4	<i>C. lutea</i>	0.44±0.01	0.51±0.01	1.85±0.16	0.175 ± 0.51
5	<i>C. dorata</i>	1.83±0.14	1.23±0.01	1.89±0.11	0.322 ± 0.40
6	<i>S. alata</i>	0.43±0.10	1.010±0.01	3.79±0.22	0.216 ± 0.33
7	<i>N. latifolia</i>	0.37±0.01	1.068±0.010	1.595±0.105	0.100 ± 0.33
8	<i>A. djalensis</i>	0.31±1.01	1.320±0.006	3.496±0.090	0.297 ± 0.32
9	<i>B. Sapida</i>	0.40±0.11	0.891±0.010	1.890±0.210	0.487 ± 0.41
10	<i>E. chlorantha</i>	0.29±0.01	0.921±0.009	5.998±0.120	0.152 ± 0.23

Values are in mean ± SEM

THE PH LEVEL AND ALKALINE PHOSPHATASE ACTIVITY IN CRUDE OIL CONTAMINATED SOIL BIOREMEDIATED WITH POULTRY MANURE AND SAWDUST

E. Jeroh ^{1*} and N. J Tonukari²

¹Biology Department, Federal Government College, P.M.B.1014, Warri, Delta State, Nigeria.

²Department of Biochemistry, Delta State University, Abraka, Nigeria.

*Corresponding author.

Email: onome4jeroh@yahoo.com and ejayetajerohbmw@yahoo.ca.

Tel: +2348059634317, +2347040060587 and +2348137936850

Abstract

The analysis of soil pH level and alkaline phosphatase activity in crude oil treated soil following remediation with poultry manure (PM) and Sawdust (SD) were studied for four weeks using standard bioremediation techniques. The result showed a statistically significant increase in soil pH level after bioremediation ($P < 0.05$) while the alkaline phosphatase activity in the soil increased upon crude oil contamination from $1.78 \pm 0.143 \text{ IU/g}$ to $1.89 \pm 0.144 \text{ IU/g}$ ($P > 0.05$). Treatment of crude oil contaminated soil with sawdust reduced alkaline phosphatase activities to $1.54 \pm 0.132 \text{ IU/g}$ on day 0 while poultry manure reduced the activity to $1.65 \pm 0.132 \text{ IU/g}$ on same day 0 ($P > 0.05$). Statistical analysis shows that only the sawdust decreased alkaline phosphatase activity significantly ($P < 0.05$) during bioremediation. The results of this study suggest that a combination of poultry manure and sawdust in bioremediation of crude oil contaminated soil is a solution to the menace of oil spillage in the Niger Delta area of Nigeria.

Key words: Crude oil contaminated soil, bioremediation, pH, alkaline Phosphatase, sawdust, poultry manure.

INTRODUCTION

Crude oil has been a major contaminant of soil and water in oil producing communities the world over during exploration and transportation and its spillage has caused critical environmental and health hazards. Crude oil spills from pipelines and refineries leads to oil pollution which causes damage to the environment (Ogbo and Okhuoya, 2008). Oil pollution is a major environmental concern in many countries and this has led to a concerted effort in studying the feasibility of using oil-degrading bacteria for bioremediation (Akoachere *et al.*, 2008). Increasing attention has been paid to developing and implementing innovative technology for cleaning up such contaminations. Bioremediation is any process that uses microorganism, fungi, green plant or their enzymes to return the environment altered by contaminants to its original or close to its original condition before contamination. The addition of organic waste material such as poultry manure, sawdust and dry leaves to the soil, facilitates aeration and increase the water holding capacity of the soil, thus enhancing bioremediation. Bioremediation is an economical and safe method for cleaning up of oil spills. Bioremediation of crude oil polluted soil is becoming increasingly important as most exploration and distribution of crude oil and its products are usually environmentally non friendly (Odokuma and Dickson, 2003).

The soil is a key component of natural ecosystem because environmental sustainability depends largely on a sustainable soil ecosystem (Adraino et al., 1998). Agricultural practices that reduce soil degradation and improve agricultural sustainability are needed particularly for tropical and sub tropical soils. Elcio *et al.*, (2004), observed that there is correlation of soil enzyme activity with total organic carbon, carbon and nitrogen biomass.

Soil enzymes (amylases, acid and alkaline phosphatases, cellulases and arylsulfatases, among others) regulate ecosystem functioning and in particular, play a key role in nutrient cycling. They catalyses certain important reactions necessary for the life processes of microorganisms in soil and the stabilization of soil structure (Makoi and Ndakidemi, 2008). Soil enzymatic activity which can be determined quite promptly and precisely is a reliable indicator reflecting the current biological state of the soil (Wyszkowska *et al.*, 2002). According to Achuba (2006), crude oil induced changes in the activities of starch degrading enzymes. Margesin and Schinner (2001), investigated the feasibility of bioremediation as a treatment option for a chronically diesel oil polluted soil in an alpine glacier area at an altitude of 2875m above sea level and observed that there was a significant reduction in the diesel oil after remediation thereby reducing the level of contamination. Measuring the success of bioremediation of oil spills is based on several parameters. This includes the degradation of polycyclic aromatic hydrocarbon (PAHs) in the crude oil (Igwo-Ezikpe, 2006). The measurement of soil enzyme activities before, during contamination and after bioremediation is used to determine the success of bioremediation (Wyszkowska and Wyszkowski, 2006).

This study, investigates the pH level and alkaline phosphatase activities of crude oil contaminated soil remediated with poultry manure and sawdust.

MATERIALS AND METHODS

MATERIALS: Soil samples were collected from a farmland in Abraka, the sawdust (SD) was collected from a sawmill in Abraka, the poultry manure (PM), from a poultry farm in Federal Government College, Warri, while the crude oil was collected from Shell Petroleum Development Company (SPDC), Warri. Other materials and apparatus used in this study included, test tubes, test tube rack measuring cylinder conical flasks, beakers, universal bottles, pH meter, spectrophotometer, laboratory coats, spatula, weighing balance, electronic digital weighing balance, micropipette, filter paper cotton wool, masking tape and plastic bowls.

METHODS: 7.3% (v/w) crude oil was added to the treated soil while the control soil had 7.3% (v/w) of distil water added to it. The experimental soil for bioremediation separated into two samples had 7.3% (v/w) crude oil. 22% (v/w) of sawdust was added to one sample while 22% (v/w) of poultry manure was added to the other. The last experimental set up had the combination of sawdust and poultry manure of 7.3 and 11.1% (v/w), respectively.

For the control sample, 10g of soil was measured into a measuring cylinder and made up to 100ml with sterile deionized water. The content was mixed and the constituent solution was filtered using Whatman No. 1 filter paper and the filtrate was kept for further analysis. The measurement of enzyme activity and the pH level were done using the filtrate solution. Alkaline phosphatase was determined by the method of Kochmar and Moss (1976), while the pH was measured using Extech pH meter.

STATISCAL ANALYSIS

The results were expressed as mean \pm SD. The one way analysis of variance (ANOVA) was used for the evaluation of statistical significance.

RESULTS

Upon contamination of soil with crude oil, the pH level of the crude oil contaminated soil (COTS) increased when compared with control soil (NC) (Table 1). Bioremediation with sawdust and poultry manure decreased the pH when compared with crude oil treated soil and normal (control) soil. Thereafter, the pH increases with time. It was observed that the remediation resulting from the combination of poultry manure and sawdust had the highest pH 7.15 ± 0.143 after 28 days of treatment of the crude oil contaminated soil. ($P < 0.05$) (Table 1).

The result from Table 2 shows that upon crude oil contamination, alkaline phosphatase activity in the soil increased. However, with bioremediation, the alkaline phosphatase activity decreased significantly ($P < 0.05$) on day 14, when the treatment of sawdust and poultry manure were combined, when compared with crude oil treated soil.

DISCUSSION

Crude oil spills from pipelines and refineries cause damage to the environment. The contamination changes the physicochemical and biological properties of the soil, as the oil is toxic to some microorganisms and plants (Ogbo and Okhuoya, 2008). Pollution of the natural environment has been observed to have adverse effects on the soil. In order to ameliorate these effects on the soil, the concept of bioremediation was initiated. Akonye and Onwudiwe (2004) reported that, the addition of organic materials such as sawdust as well as dry grass helped in the remediation process of the soil. Adedokun and Ataga (2007) also reported that for efficient bioremediation, soil amendments or additives such as sawdust, peat, waste cotton manure, fertilizers could be added to increase activities of microorganisms. Wyszowska *et al.*, (2002) stated that, the activity of alkaline phosphatase was dependent on the experimental series and degree of soil contamination with diesel oil.

The results from this investigation show that upon contamination of the soil, there was an increase in the pH level ($p < 0.05$). This finding is in agreement with Adenipekun (2008), who observed that contamination of soil with crude oil increased pH level. The result also shows that when treated with soil was remediated with poultry manure and sawdust; the contaminated soil regained its original status. The level of alkaline phosphatase activity in the soil increased upon crude oil contamination but was observed to reduce also after remediation with poultry manure and sawdust.

The use of poultry manure in the treatment of crude oil contaminated soil was observed to significantly increase phosphatase activity (Table 2). This increase in phosphatase activity may be due to the ability of the sawdust present in poultry manure to absorb the oil films thereby reducing acid radicals released into the soil; thus, reducing the toxicity effect of the contaminant. This result may be added to the fact that increase in phosphorus activity is due to the addition of poultry manure (PM), which mobilizes microorganism that secretes phosphatase, to hydrolyse phosphorus. These findings are in agreement with Wyszowska and Wyszowski (2006), who stated that that diesel oil stimulated the activity of dehydrogenases, ureases, alkaline phosphatase as well as nitrification, but inhibited the activity of acid phosphatase.

CONCLUSION

This present research has shown that pH level of soil increased upon oil contamination. This is expected since crude oil is alkaline in nature. However, crude oil contamination of soil over a long time may lead to a drop in pH due to the release of hydrogen ions in the soil which will thus lead to soil toxicity. Such environment, if bioremediated with poultry manure will return such soil close or back to its original status before contamination took place. Alkaline phosphatase activity was increased upon oil contamination. Treatment of such soil with sawdust helped in reducing the

alkaline phosphatase activity. The results of this study clearly showed that a combination of poultry manure and sawdust can be used as a good bioremediation material on a crude oil contaminated soil.

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Table 1. The pH levels of crude oil contaminated soil treated with poultry manure and sawdust.

Samples	Day 0(mean±sd)	Day 14(mean±sd)	Day28(mean±sd)
Normal control soil	6.64 ± 0.123b	6.40 ± 0.122b	0.123b 6.37 ±
Crude oil treated soil (COTS)	7.76 ± 0.142a	6.75 ± 0.134b	0.144b 6.70 ±
COTS + sawdust (SD)	6.37 ± 0.122b	6.59 ± 0.125b	0.142a 7.07 ±
COTS + poultry manure (PM)	6.56 ± 0.125b	6.84 ± 0.128b	0.132b 6.94 ±
COTS + PM + SD	6.45 ± 0.122b	6.90 ± 0.127b	0.143a 7.15 ±

Table 2. Alkaline phosphatase activities of crude oil contaminated soil treated with poultry manure and sawdust.

Samples	Day 0(mean±sd)	Day 14(mean±sd)	Day28(mean±sd)
Normal control soil	1.78 ± 0.143a	1.71 ± 0.143a	1.71 ± 0.132a
Crude oil treated soil (COTS)	1.89 ± 0.144a	1.88 ± 0.132a	1.98 ± 0.135a
COTS + sawdust (SD)	1.54 ± 0.132b	1.54 ± 0.133b	1.54 ± 0.135b
COTS + poultry manure (PM)	1.65 ± 0.133a	1.75 ± 0.133a	1.81 ± 0.144a
COTS + PM + SD	1.42 ± 0.125b	1.54 ± 0.125b	1.64 ± 0.142a